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**Helminth Infection Does Not Reduce Risk for Chronic Inflammatory Disease in a
Population-Based Cohort Study**

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ABSTRACT

Background & Aims: Parasitic helminth infections can suppress symptoms of allergy, type I diabetes, arthritis, and inflammatory bowel disease in animal models. We analyzed data from a large, population-based cohort study to determine whether common childhood enterobiasis protects against these diseases.

Methods: We collected information on individual prescriptions filled for the drug Mebendazole against *Enterobius vermicularis* for all children born in Denmark 1995–2008 from the National Register of Medicinal Product Statistics (n=924,749; age 0–14 years); 132,383 of these children (14%) filled a prescription for Mebendazole, 102,482 of the children (11%) had a household peer who was registered with a filled Mebendazole prescription, and the remaining 689,884 children (75%) comprised the reference group. Children diagnosed with asthma, type I diabetes, juvenile arthritis, ulcerative colitis, or Crohn's disease were identified from the National Patient Registry. We used Poisson regression to estimate confounder-adjusted incidence rate ratios (IRRs) for first in- or out-patient hospital diagnosis of chronic inflammatory disease according to history of Mebendazole treatment prescribed to children in the study.

Results: Chronic inflammatory disease was diagnosed in 10,352 children during 6.4 million person-years of follow-up. The IRR was 1.07 for asthma (95% confidence interval (CI), 1.00–1.13), 1.05 for type I diabetes (95% CI, 0.79–1.12), 1.13 for juvenile arthritis (95% CI, 0.94–1.37), 0.77 for ulcerative colitis (95% CI, 0.41–1.46), and 1.44 for Crohn's disease (95% CI, 0.82–2.53). The results were not modified by number of treatments or age at treatment.

Conclusion: Based on a population-based analysis, enterobiasis does not reduce the risk for asthma, type I diabetes, arthritis, or inflammatory bowel disease.

Keywords: parasite infection; immune response; inflammation; risk factor

INTRODUCTION

Many high-income countries have experienced increases in the occurrence of a diverse range of chronic diseases characterized by immune reactions to otherwise harmless antigens. An intriguing hypothesis is that components of the immune system may have evolved to “silence” parasitic helminth (worm) infections and has gone awry in their absence.¹⁻⁴ In a few clinical trials, reintroduction of helminth exposure was shown to be effective therapy for inflammatory bowel disease,^{5, 6} but not for asthma or allergic rhinitis.^{7, 8} More studies support the idea that helminth exposure might protect against the development of chronic inflammatory disease. For instance, helminths administered *before* disease induction suppress disease activity across rodent models for allergic diseases,⁹⁻¹² type I diabetes,¹³⁻¹⁹ arthritis,²⁰⁻²³ inflammatory bowel disease,²⁴⁻²⁸ and multiple sclerosis,²⁹⁻³³ with few exceptions.³⁴⁻³⁸ Furthermore, epidemiological studies, mostly of children, have reported that current infection with certain helminths, e.g. hookworms and schistosomes, is associated with a consistent reduction in risk of asthma³⁹ or sensitization to allergens.⁴⁰ However, other chronic inflammatory diseases have not been studied, and the role of enterobiasis (pinworm) has received little attention *á priori*.⁴¹⁻⁴⁴ *Enterobius vermicularis* has the largest geographical distribution among helminths and still infects up to 20% of children in many high-income countries.^{45, 46} Treatment with the first-choice drug Mebendazole is common, and can be used as an indicator for having enterobiasis. Using the national drug and patient registries in Denmark, we therefore observed a nationwide cohort of children and determined the risk of a range of chronic inflammatory diseases after use of Mebendazole.

MATERIALS AND METHODS

Study cohort and data sources

A cohort of 924,749 children born alive in Denmark during the period 1 January 1995 through 31 December 2008 was established using the Civil Registration System,⁴⁷ and subsequently linked with the Register of Medicinal Product Statistics, the Integrated Database for Labor Market Research, and the National Patient Registry. Accurate person linkage between registers is possible because all information in national registers is kept under the unique Danish personal identification number assigned to all Danish residents since 1 April 1968.⁴⁷ Since January 1, 1995, the Register of Medicinal Product Statistics has contained monthly updated information on all persons' filled prescriptions for subsidized and non-subsidized medicinal products that have a marketing authorization in Denmark (www.dkma.dk).⁴⁸ The information has a high national coverage because prescribed medicine can be collected/purchased at pharmacies only and all pharmacies have a duty by law to report information to the register. Information on, e.g., number of dispensed packages of a product, Danish medicinal product code on labels of dispensed packages, and updated international Anatomic-therapeutic Chemical (ATC) code associated with the Danish product code, is kept for every filling date linked to the personal identification number recorded on the prescription. The Integrated Database for Labor Market Research contains longitudinal information for the entire Danish population about, e.g., total income and number of persons living in each household from 1980 to the present. The National Patient Registry contains information about all non-psychiatric hospital admissions in Denmark since January, 1977.⁴⁹ Outpatient hospital contacts have been included since 1995. Information concerning dates of admission and discharge, hospital department and diagnoses is kept for every

hospital contact linked to the patient's personal identification number. Diagnoses are coded according to the 8th revision of the *International Classification of Diseases* (ICD) since January 1, 1977, and the 10th revision since January 1, 1994. The study was based on existing data and approved by the Danish Data Protection Agency (j.nr. 2008-54-0472) and the Danish Medicine Agency. Because subjects were not contacted during the study, written informed consent was not required.

Anthelmintic treatment

From the Register of Medicinal Product Statistics we obtained information on all filling dates between 1995 and 2008 for Mebendazole prescriptions (ATC code PO2CA01, Danish tradename "Vermox") issued to persons in Denmark. We considered the date of filling a prescription (i.e. purchasing Mebendazole) as the approximate date of use of Mebendazole against enterobiasis.

Chronic inflammatory disease

Chronic diseases included in previous animal models cited in the introduction section, and without rare onset and hospitalization in childhood, were selected. For each disease, in- and outpatient cases were identified in the National Patient Registry using ICD-10 codes: Asthma (J45, J46), type I diabetes (E10), juvenile arthritis (M080, M082, M083, M084, M088, M089), ulcerative colitis (K51) and Crohn's disease (K50). For asthma, diagnoses before the age of 5 years were excluded due to the risk of misclassification of asthma-like symptoms with a different etiology (e.g., early childhood infection).

Potential confounder variables

Variables based on the Civil Registration System included age (0, 1, 2, ..., 14 years), sex, calendar period (1995-99, 2000-05, 2005-08), birth order (1 [for firstborn], 2, and ≥ 3), degree of urbanization (<25 , 25-349, 350-999, 1000-1999, ≥ 2000 persons/km²), birth place of parents (one or both born in Denmark, both born abroad), maternal age (≤ 19 , 20-24, 25-29, 30-34, ≥ 35 years), and number of household peers (1, 2, 3, ≥ 4 persons). Household peers (parents, parental partner, guardians, siblings, half-siblings, step-siblings) were defined by an algorithm using information on addresses, sibling relations, marital status, sex, and age for all persons alive and living in Denmark since 1995. The variable was designed by Statistics Denmark (www.ds.dk). A variable for average gross annual income for each individual was based on information in the Integrated Database for Labor Market research, and calculated as household income divided by number of adults in the household, scaled with a price index to make the income comparable over the study period, and then grouped into 5 levels of 100,000 Danish kroner (DKK) ($\sim 10,500$ Great Britain Pounds $\sim 21,000$ US Dollars in 2008), with the highest level being $>500,000$ DKK. A variable denoting parental chronic inflammatory disease status, i.e. the same type of disease in one or both biological parents (yes/no) was based on information in the National Patient Registry: Again ICD-10, but also the ICD-8 codes shown in the following parentheses, were used, and we hereby identified parents with any diagnosis of asthma (493.00-493.02, 493.08-493.09) between the age of 5 and 50 years, type I diabetes (249.00-249.09), juvenile arthritis (712.09), ulcerative colitis (563.19, 569.04) or Crohn's disease (563.00-563.02, 563.08-563.09). Finally, the following variables were treated as time-dependent: Age, calendar period, degree of urbanization, average gross annual income, and parental chronic inflammatory disease.

Statistical analyses

Each child was followed from birth until first in- or outpatient hospital contact for chronic inflammatory disease, death, emigration, or 31 December, 2008, whichever came first. For asthma as single outcome, children were followed from the age of 5 year only. Log-linear Poisson regression models were used to estimate competing incidence rate ratios (IRRs) for chronic inflammatory disease by history of filled Mebendazole prescription. A history of filled Mebendazole prescription was treated as a time-varying variable with the categories “Yes”, “No”, or “Other”: The category “Yes” consisted of children registered with a filled Mebendazole prescription issued in their name, i.e. personal registration number. In the category “Other” we included children who had no Mebendazole prescription but had a household peer who had. In this way, the reference category “No” consisted of the remaining children with no Mebendazole prescription themselves or for any household peer. Finally, the category “Yes” were further divided according to the three time-varying variables related to Mebendazole prescriptions: Time since latest prescription, age at first prescription, and number of prior filled prescriptions. We chose the first variable to study effects indicating that intestinal symptoms of enterobiasis and inflammatory bowel disease were interchanged by mistake,⁵⁰ the second to study effects of immunological maturity, and the third to study effects of heavy/persistent enterobiasis or reinfections.

IRRs were adjusted for age, sex, and calendar period as well as all variables that were significantly ($P < 0.05$) associated with any of the five disease outcomes in initial multivariable models including all potential confounder variables; excluded were

maternal age and number of household peers. Maximum likelihood estimation of IRRs and 95% confidence intervals (CIs) was performed using the GENMOD procedure in SAS software, version 9.2 (SAS institute Inc., Cary, North Carolina). To evaluate the effect of the modelling of the variance, two alternative approaches were used. The approaches were applied on a 20% random sample of the cohort. In one approach a sandwich estimator taking into account correlation within families was used to model the variance (SAS procedure GENMOD), and in a second approach we used a random effect model with family as the random effect (SAS procedure NLMIXED). Both approaches produced results similar to the traditional Poisson regression model for the main analysis of each disease outcome (yes vs. no). We therefore presented all results based on the traditional Poisson regression model. Two-sided p-values were based on likelihood ratio tests, and 95% CIs on Wald's statistics.

RESULTS

The cohort included 924,749 children, age 0 to 14 years, of whom 132,383 (14%) children were registered with a filled Mebendazole prescription issued in their name. Among children with no filled Mebendazole prescription issued in their name, 102,482 (11%) had a household peer who had a Mebendazole prescription (group denoted "Other" in the tables), and the remaining 689,884 (75%) reference children had no such household peer. Overall, 10,352 children (1.11%, or 162/100,000 per year) developed a chronic inflammatory disease during 6,405,013 person-years of follow-up between 1995 and 2008.

Table I presents the IRR for asthma, type I diabetes, and juvenile arthritis by history of filled Mebendazole prescription (i.e. use of Mebendazole). Overall, use of Mebendazole (i.e. indicating enterobiasis) was associated with a small and significant increase of 5% in the IRR for having any of the three diseases (IRR, 1.05; 95% CI, 1.00-1.12) when compared with non-users. For each disease, there was a small increase of 5-13% in the respective IRRs, which was significant for asthma ($P=0.04$) and not type I diabetes ($P=0.45$) and juvenile arthritis ($P=0.23$). Among the Mebendazole users, neither age at first filled Mebendazole prescription ($P_{\text{any}}=0.21$, $P=0.70$, $P=0.37$, and $P=0.33$), nor prior number of filled Mebendazole prescriptions ($P_{\text{any}}=0.22$, $P=0.45$, $P=0.41$, and $P=0.54$) was associated with the IRR of having any disease, asthma, type I diabetes, and juvenile arthritis, respectively (see table I).

Table II presents the IRR for inflammatory bowel disease by history of filled Mebendazole prescription. Overall, there was no significant association between use of Mebendazole and inflammatory bowel disease (IRR, 1.33; 95% CI, 0.97-1.83) when compared with reference children. However, in strata compared with reference children, the IRR was significantly increased for children with 0-1 years (IRR 1.77; 95% CI, 1.17-2.67) but not 2-14 years (IRR 1.06; 95% CI, 0.70-1.62) since latest filled Mebendazole prescription. Correspondingly stratified IRRs for the combined outcome in table I was close to unity (IRR_{0-1y} 1.08; 95% CI, 1.00-1.17, IRR_{2-14y} 1.03; 95% CI, 0.96-1.11). The short period of 0-1 years suggested a bias, e.g., some GPs might have misclassified reports of beginning symptoms of ulcerative colitis with enterobiasis (proctitis with pruritus anis) and prescribed Mebendazole. Therefore, further analyses were restricted to users with 2-14 years since latest filled Mebendazole prescription.

The results of the analyses, presented in table II, show that neither age at first ($P=0.60$) nor prior number of ($P_{\text{intestinal}}=0.28$) filled Mebendazole prescriptions was associated with the IRR for inflammatory bowel disease.

In one robustness analyses, in which outcomes were restricted to cases with two hospital contacts for chronic inflammatory disease, there was again no association between a history of a filled Mebendazole prescription and any asthma, type I diabetes, and juvenile arthritis: IRR_{0-1y} , 0.99 (95% CI 0.90-1.10), IRR_{2-14y} 0.98 (95% CI 0.90-1.08), IRR_{Other} , 1.02 (95% CI 0.95-1.10), or any inflammatory bowel disease: IRR_{0-1y} , 1.29 (95% CI 0.65-2.58), IRR_{2-14y} 1.22 (95% CI 0.72-2.06), IRR_{Other} 1.27 (95% CI 0.79-2.05). In another robustness analysis, IRRs were calculated among children with no siblings and again the results were robust for asthma, type I diabetes, and juvenile arthritis: IRR_{0-1y} 1.01 (95% CI 0.89 - 1.15), IRR_{2-14y} 1.17 (95% CI 0.71-1.91), IRR_{Other} 1.00 (95% CI 0.94 - 1.06), as well as for inflammatory bowel disease, although here the number of cases who were prescribed Mebendazole was small: IRR_{0-1y} 1.04 (95% CI 0.46-2.37; 6 exposed cases), IRR_{2-14y} (no exposed cases), IRR_{Other} 1.37 (95% CI 0.98-1.92).

DISCUSSION

This nationwide study of Danish children does not support the hypothesis that enterobiasis protects against development of chronic inflammatory disease - even if acquired early in life or several times.

The study had a strong statistical power that relied on the immense size of our national cohort, and does not support the existence of even small protective effects. For example, based on 95% confidence intervals, the highest risk reduction compatible with our data was 0% for asthma, 21% for type I diabetes, 6% for juvenile arthritis, and 30% for inflammatory bowel disease. In comparison, a meta-analysis of 33 studies of helminths and asthma found that current infection with hookworm is associated with a 50% reduction in the risk of asthma (95% CI 10-72%), enterobiasis an insignificant risk reduction of 13% (OR 0.87, 95% CI 0.43-1.75),^{42, 43, 51-56} and *Ascaris lumbricoides* a risk increase of 34% (95% CI 5-71%).³⁹ The latter risk increase has been attributed to an asthma-like eosinophil pneumonitis, Loeffler's syndrome, provoked by ascarid larvae migrating through the lungs.^{57, 58} Another meta-analysis of 21 studies found that current infection with hookworms, *A. lumbricoides*, *Trichuris trichiura*, or schistosomes was associated with an overall reduction of 38% (95% CI 24-40%) in the risk of having skin-reactivity to at least one allergen, although not house dust mite and cockroach - allergens that typically trigger asthma attacks.⁴⁰

The cited meta-analyses could make restricted conclusions about causality, because no prospective cohort studies were found in the systematic literature search, only cross-sectional studies and case-control studies, and such studies may be prone to bias. To our knowledge the present study is the first prospective cohort study addressing a relationship between helminth infection and asthma. In the cited meta-analyses, bias was reduced by focusing on effects of stool-tested *current* helminth infection, but this restricted the conclusions to concern effects on established asthma and skin-reactivity to allergens. Such effects, however, are not supported by recent clinical trials in which

controlled hookworm or pig whipworm (*Trichuris suis*) infection demonstrated no effectiveness against asthma,⁷ skin-reactivity to allergens,^{7, 59} or allergic rhinitis.⁸ Considering these inconsistent results, a causal explanation for the protective effects found in the meta-analyses could be that helminth infection suppress the *short priming* with allergen, rather than the subsequent *chronic reactivity* to allergen which progresses clinical disease.⁶⁰ Other poorly addressed explanations are, e.g., reverse causation whereby allergic individuals are less likely to establish helminth infections, or a bias, e.g., from unintended exclusion of infected allergic individuals from some research projects.

Very few previous studies investigated an association with enterobiasis *á priori* by using perianal tape test,⁴²⁻⁴⁴ whereas others^{51-56, 61} lacked statistical power because they used stool samples and were smaller studies.^{39, 45} For example, prevalences of enterobiasis among *children* were 14-29% using perianal tape test,⁴²⁻⁴⁴ and 0-8% using stool samples.^{52-56, 61} One *á priori* study reported an association between current enterobiasis and asthma,⁴² while the remaining studies reported neither an association with asthma^{43, 44, 51-56} nor with skin-reactivity to allergens.^{44, 61} The former *á priori* study used school records and parental questionnaires to recall the result of routine tape test screening among 3,107 school children in a Taiwanese city, and reported a 75% (95% CI 37-90) reduction in risk of parent-reported doctor-diagnosed current asthma, and no association with atopic dermatitis or parental allergy.⁴²

In general, the lack of association with enterobiasis reported in most previous studies^{43, 44, 51-56} is in agreement with our results for asthma, although lack of statistical power is a

more likely explanation for most previous findings. Our large national cohort allowed us to identify a small 7% (0-13%) excess risk of asthma. However, this finding should be interpreted with caution. The small size of the effects in the confidence intervals is difficult to make compatible with a clinically relevant susceptibility towards asthma among children who had enterobiasis, and can easily be due to small biases.

By using Mebendazole prescriptions as a proxy for enterobiasis, the existence of the Danish drug and patient registries allowed us to test the suggested hypothesis in an entire population and in a prospective follow-up design. The strength of this method derived from the following reasons that enterobiasis and chronic inflammatory disease would unlikely or rarely be misclassified:

We used identification of filled prescriptions for Mebendazole in a register that covers the entire Danish population over the last 15 years. By using the filling dates as a proxy for time of enterobiasis, we avoided recall bias. We feel confident that most Mebendazole prescriptions are filled, because untreated enterobiasis may persist in a family for months despite hygienic precautions, and treatment with Mebendazole is inexpensive and safe. To assess potential misclassifications, we initially considered how well children *with* or *without* enterobiasis would be classified when using their Mebendazole prescription as a proxy for enterobiasis. A correct classification of children *with* enterobiasis is likely determined by three factors. First, our experience is that to control spread^{45, 46} then most GPs in Denmark will prescribe Mebendazole based on signs of enterobiasis. Because these signs are quite unique (e.g., persistent nocturnal anal pruritus) then misclassification would be rare.⁶² Second, a number of GPs might

recommend Pyrvinium (Danish tradename “Vanquin”) instead of Mebendazole, although it discolors with red and less is known about side-effects. Because Pyrvinium is a non-prescription drug (i.e. over-the-counter), the Register of Medicinal Product Statistics receives information about packages purchased at pharmacies *and* groceries (i.e. both have a duty by law to report to the register) – but by product code only, not by person. However, with that information we could total the purchase of Pyrvinium and Mebendazole and estimate that only 2% of our reference children used Pyrvinium - a fraction unlikely to have biased our study results toward unity. Third, some GPs might prescribe *excess* Mebendazole packages to one infected child, e.g., as a convenient way to provide the household with enough Mebendazole to treat all its members. In this way however, other *infected* children in the household would be misclassified. We avoided this potential misclassification by grouping all children who had household peers with a prescription for Mebendazole separately (the group “Other” in the tables). Finally, correct classification of reference children *without* enterobiasis would depend mainly on two factors. First, Mebendazole can be prescribed for other nematodal helminth infections (e.g., *Ascaris* and *Trichiura* species), however because such infections are rare in Denmark then the misclassification would be rare.⁶³ Second, to control spread some GPs might prescribe Mebendazole to each and every household member, even to siblings who were not infected. However, when restricting our analyses to children without siblings, our results were robust.

We used as outcome national hospital diagnoses with a high validity and completeness for inflammatory bowel disease (90-97% and 94%), type I diabetes (96% and 91%), and juvenile arthritis.^{64, 65} We increased the validity of the asthma diagnoses by restricting

diagnoses to ages 5-14 years, but we did not identify all asthmatic children since many have had no hospital contact at this age.⁶⁶ However, we observed that the lack of effect was robust for children who had two hospital contacts for chronic inflammatory disease including asthma. Potential misclassification of chronic inflammatory disease is therefore most likely not to explain our null results.

A limitation of our study was that we could not outrule some GPs mistakenly prescribed Mebendazole for intestinal symptoms of as yet undiagnosed inflammatory bowel disease. We were inclined to believe in such bias, because we observed an excess risk of being diagnosed with ulcerative colitis in the two years after latest prescription but not later (see sublegend in table II, $P=0.002$), and in previous studies using the same national registers, there was excess diagnostic activity (i.e. an uncertainty) and prescription of antibiotics in the year before diagnoses of inflammatory bowel disease.^{67, 68} The alternative explanation, that enterobiasis somehow trigger ulcerative colitis would obviously need confirmation in future studies, and is incompatible with the rest of our results, as well as most experiments with helminths and colitis²⁴⁻²⁸ although not all.³⁶

Interestingly, enterobiasis and other helminths that dwell in the intestine have been reported to induce *local* immune-suppressive T cells and cytokines in inflammatory bowel disease patients, and to reduce disease activity in some cases.^{5, 6, 69-71} In addition, experimental studies have identified a number of immune-suppressive properties of helminth-derived products.⁷² Our population-based observations suggest that enterobiasis can neither be immune-suppressive nor balance early immune maturation,

in a way that protect against diseases with chronic reactions to otherwise harmless antigens in the intestine, pancreas, joints, or lungs. It might be argued that enterobiasis is a short-term infection of weeks to months, whereas prolonged (years) or repeated helminth infections, as occur in many low-income countries, would be more likely to exert protective effects. However, we found no evidence that repeated enterobiasis protect against the studied chronic inflammatory diseases.

In contrast to pinworms (enterobiasis), hookworms and schistosomes thrive in *tropical* regions only. Because increases in the occurrence of chronic inflammatory disease mainly have occurred in *non-tropical* countries, i.e. mainly high-income countries, then it is not logical that an absence of tropical helminths is an explanation. The present study was conducted in a high-income country and does not support that a common helminth like enterobiasis protects against chronic inflammatory disease among children. Taken together, we believe one possible explanation could be that absence of helminths has had little impact on the increase in the incidence of chronic inflammatory diseases in high-income countries.

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Table I. Incidence rate ratios for three chronic inflammatory diseases according to history of filled individual-level prescriptions for the first-choice drug Mebendazole against enterobiasis in a cohort of all children born in Denmark 1995-2008.

Any disease				Asthma*		Type I diabetes		Juvenile Arthritis	
Cases	PYRS	IRR (95% CI)		Cases	IRR (95% CI)	Cases	IRR (95% CI) [§]	Cases	IRR (95% CI)
Filled prescription for Mebendazole									
No	7,183	5,265	1 (reference)	5,334	1 (reference)	952	1 (reference)	897	1 (reference)
Yes [†]	1,630	609	1.05 (1.00, 1.12) [‡]	1,349	1.07 (1.00, 1.13) [‡]	135	1.05 (0.79, 1.12) [‡]	132	1.13 (0.94, 1.37) [‡]
Age (years) at first filled prescription									
0-2	455	218	1.06 (0.97, 1.17)	362	1.08 (0.97, 1.20)	54	1.09 (0.83, 1.44)	39	0.93 (0.68, 1.29)
3-5	865	298	1.04 (0.97, 1.11)	730	1.05 (0.97, 1.13)	70	0.89 (0.69, 1.14)	65	1.21 (0.94, 1.58)
6-8	258	79	1.07 (0.94, 1.21)	216	1.08 (0.94, 1.24)	18	0.72 (0.45, 1.15)	24	1.48 (0.97, 2.25)
9-14	52	15	1.22 (0.93, 1.61)	41	1.26 (0.92, 1.72)	7	1.22 (0.57, 2.60)	4	0.95 (0.35, 2.58)
Prior number of filled prescriptions (i.e. prior to the latest) ^{††}									
0	1,104	433	1.04 (0.98, 1.11)	914	1.05 (0.98, 1.13)	98	0.90 (0.73, 1.11)	92	1.13 (0.91, 1.41)
1	293	106	1.03 (0.91, 1.15)	247	1.06 (0.93, 1.20)	26	0.90 (0.61, 1.34)	20	0.98 (0.62, 1.53)
2+	233	70	1.18 (1.03, 1.34)	188	1.16 (1.01, 1.35)	25	1.22 (0.88, 1.82)	20	1.39 (0.88, 2.18)
Other ^{‡‡}	1,197	531	1.00 (0.94, 1.06) [‡]	948	0.98 (0.91, 1.05)	135	1.05 (0.87, 1.27)	114	1.13 (0.93, 1.39)

IRR, competing incidence rate ratios; PYRS, person-years at risk (in thousands); CI, confidence intervals. IRRs and CIs are estimated in a traditional Poisson regression model with adjustment for age, sex, calendar period, birth order, average gross annual income based on the total household income, degree of urbanization, birth place of parents, and any parental diagnoses of the specific outcome.

* Excluding diagnosis of asthma before the age of 5 years.

† The IRRs did not change materially with different modeling of variance (traditional Poisson regression, p; Poisson regression with variance modeled by a sandwich estimator, s; Poisson regression with a random family effect, r) in a random 20% sample: Any disease: IRR_p 1.16 (0.99-1.35), IRR_s 1.16 (0.99-1.35), IRR_r 1.16 (0.99-1.35); Asthma: IRR_p 1.19 (1.00-1.41), IRR_s 1.19 (1.00-1.41), IRR_r 1.19 (1.00-1.41); Type I diabetes: IRR_p 0.98 (0.58-1.67), IRR_s 0.98 (0.58-1.67), IRR_r 0.99 (0.58-1.67), Juvenile Arthritis: IRR_p 1.23 (0.73-2.08), IRR_s 1.23 (0.73-2.09), IRR_r 1.23 (0.73-2.09).

‡ The IRRs did not change materially: When adjusted for age, sex, and calendar period only: IRR 1.04 (0.98-1.09), IRR_{other} 0.97 (0.91-1.03). When stratified by time since latest filled prescription (see also table II): P=0.40, IRR_{0-1y} 1.08 (1.00-1.17), IRR_{2-14y} 1.03 (0.96-1.11). When further adjusting for parental inflammatory bowel disease.

†† Multiple prescriptions were defined as those filled >4 weeks apart, and were considered a proxy for heavy/persistent infections or reinfections. Using intervals of >12 weeks did not change the results materially. The recommended treatment duration using Mebendazole tablets or mixture was 5-8 weeks for normal infections (100 mg as single dose for a total of 3 times with 2-3 weeks interval), and 11 weeks for heavy infections (100 mg as single dose for a total of 6 times with 2 week intervals).

‡‡ Includes children who had not themselves had a Mebendazole prescription but who lived in a household with a person who had (e.g., a sibling or parent).

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Table II. Incidence rate ratios for inflammatory bowel disease according to history of filled individual-level prescriptions for the first-choice drug Mebendazole against enterobiasis in a cohort of all children born in Denmark 1995-2008.

Inflammatory bowel disease			
Cases	PYRS	IRR (95% CI)	
Filled prescription for Mebendazole			
No	245	5,265	1 (reference)
Yes	53	609	1.33 (0.97, 1.83)*
P=0.07			
Time since latest filled prescription (years)			
0-1	26	308	1.77 (1.17, 2.67) [†]
2-14	27	301	1.06 (0.70, 1.62)
P=0.07 [‡]			
Age (years) at first filled prescription			
0-2	7	120	0.90 (0.42, 1.93)
3-5	18	147	1.13 (0.70, 1.83)
6-8	2	31	-
9-14	0	3	-
P=0.60			
Prior number of filled prescriptions ^{††}			
0	22	233	1.19 (0.76, 1.88)
1	3	46	0.71 (0.29, 1.75)
2+	2	22	-
P=0.28			
Other ^{††}	44	531	1.36 (0.97, 1.90)

IRR, competing incidence rate ratios; PYRS, person-years at risk (in thousands); CI, confidence interval. IRRs are adjusted for age, sex, calendar period, birth order, average gross annual income based on the total household income, degree of urbanization, birth place of parents, and any parental inflammatory bowel disease.

* The IRRs did not change materially when adjusting for age, sex, and calendar period only: IRR 1.34 (0.98-1.83), IRR_{other} 1.36 (0.97-1.89).

† Time was categorized in 0-1 and 2-14 years because of similar direction of IRR when using detailed intervals, e.g., IRR_{0y}=1.76, IRR_{1y}=1.77, IRR_{2y}=0.20, IRR_{3y}=1.19, and IRR_{4y}=0.53.

‡ Ulcerative colitis: P=0.002, IRR_{0-1y} 2.39 (1.48-3.87), IRR_{2-14y} 0.77 (0.41-1.46), IRR_{other} 1.28 (0.81-2.01). Crohn's disease: P=0.37, IRR_{0-1y} 0.94 (0.41-2.17), IRR_{2-14y} 1.44 (0.82-2.53), IRR_{other} 1.45 (0.88-2.40). Because we could not outrule a non-causal association for the strata 0-1 year (GPs could have misclassified beginning symptoms of ulcerative colitis with enterobiasis, e.g., proctitis with pruritus anis, and prescribed Mebendazole) further analyses below in the

table were restricted to children in the strata 2-14 years.

†† Multiple prescriptions were defined as those redeemed >4 weeks apart, and were considered a proxy for heavy/persistent infections or reinfections. See also table I.

‡‡ Includes children who had not themselves had a Mebendazole prescription but who lived in a household with a person who had (e.g., a sibling or parent).