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Age-associated carriage of intestinal parasites among adults in north-eastern Poland – a cross-sectional study

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Abstract

Introduction and Objective. Data concerning the occurrence of intestinal parasites among the Polish population remain limited. The aim of the study is to assess the carriage of intestinal parasites among adults from north-eastern Poland and to identify demographic and clinical factors associated with their detection.

Materials and Methods. A cross-sectional study was conducted among 118 adults from the Podlaskie Province in north-eastern Poland. Stool samples were examined microscopically for the presence of intestinal parasites and other intestinal eukaryotic organisms. In addition, molecular assays targetting selected clinically relevant intestinal parasites were performed. Demographic characteristics and gastrointestinal symptoms were collected using a structured questionnaire. Associations between organism detection and demographic or clinical variables were assessed statistically.

Results. Intestinal organisms were detected in 20/118 participants (16.9%), including 19 (16.1%) harbouring pathogenic or potentially pathogenic species. The most frequently identified organisms were *Blastocystis* spp. (n = 13) and *Dientamoeba fragilis* (n = 5). Single cases of *Giardia intestinalis*, *Enterobius vermicularis*, *Entamoeba coli*, and *Endolimax nana* were observed. Increasing age was significantly associated with the presence of intestinal organisms (p = 0.016).

Conclusions. The prevalence of intestinal parasites among adults in north-eastern Poland was low and was largely driven by the detection of *Blastocystis* spp. and *Dientamoeba fragilis*. Pathogenic parasites were rare, whereas older age was associated with a higher likelihood of intestinal organism detection. Intestinal parasites, particularly *Blastocystis* spp. and intestinal protists, remain an important component of adult gut ecology and epidemiology despite the low burden of clinically significant parasitic infections.

Key words

adults, Poland, intestinal parasites, *Blastocystis* spp., *Dientamoeba fragilis*

INTRODUCTION

Intestinal parasitic infections and the presence of other intestinal eukaryotic organisms remain an important component of parasitological diagnostics, including in countries with a high sanitary standard [1, 2]. Although the overall prevalence of intestinal parasitic infections has declined substantially in Europe, both pathogenic parasites and intestinal eukaryotes of uncertain clinical significance continue to be detected in adult populations [2, 3].

Parasitological testing in adults is still performed predominantly in symptomatic individuals, which may contribute to underestimation of the true prevalence of intestinal infections and colonization [4]. In particular, *Giardia intestinalis*, despite its well-established pathogenic potential, may occur asymptomatically and therefore remain undiagnosed in the absence of routine stool examination [5].

In recent years, increasing attention has been paid to intestinal eukaryotes, such as *Blastocystis* spp., *Dientamoeba fragilis*, *Endolimax nana*, and *Entamoeba coli*. While the latter species are generally regarded as non-pathogenic indicators of faecal–oral exposure, the clinical relevance of *Blastocystis* spp. and *D. fragilis* remains controversial [6–8]. Furthermore, several European studies have reported age-related differences in the prevalence of intestinal eukaryotes, with higher detection rates observed among older individuals [9, 10].

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The aim of the study was to assess the carriage of intestinal parasites and other intestinal eukaryotic organisms among adults from north-eastern Poland and to evaluate demographic and clinical factors associated with their detection, with particular emphasis on the relationship between age and the presence of intestinal organisms.

MATERIALS AND METHODS

Study design and population. A cross-sectional study was conducted between October 2024 – February 2025 among residents of the Podlaskie Province in north-eastern Poland (Fig. 1). Participants were recruited consecutively on a voluntary basis, regardless of gender or the presence of gastrointestinal symptoms. Eligible individuals were aged ≥18 years and resided in the Podlaskie. Written informed consent was obtained from all participants prior to enrollment. A total of 118 adults were included in the study.

Figure 1. Geographic location of the study area in Europe. Poland is outlined in dark grey, and the Podlaskie Province highlighted in blue

Data collection and sample processing. Participants completed a structured questionnaire collecting socio-demographic data and gastrointestinal symptoms. Each participant provided three stool samples collected on separate days. Two samples were preserved in sodium acetate–acetic acid–formalin (SAF) solution for microscopic examination, while one sample was preserved in 96% ethanol for molecular analysis.

Microscopic examination. Two SAF-preserved stool samples per participant (236 samples in total) were examined using direct wet mount microscopy with Lugol's iodine, decantation in distilled water, and Fülleborn flotation techniques. All slides were independently evaluated by three experienced laboratory diagnosticians. Microscopy served as the primary diagnostic method for the detection of intestinal parasites.

Molecular analysis. DNA was extracted from ethanol-preserved stool samples (n = 118) using the Bosphore Versatile DNA Isolation Kit (Anatolia Geneworks, Turkey) according to the manufacturer's instructions. Prior to extraction, samples underwent ethanol removal, washing, thermal shock cycles, and mechanical disruption.

Real-time PCR was performed using the Bosphore Parasitic GI Panel (Anatolia Geneworks, Turkey), targeting *Ascaris* spp., *Taenia* spp., Enterobius vermicularis, *Entamoeba histolytica*, *Cryptosporidium* spp., and *Giardia duodenalis*. Amplification was carried out on an AriaMx Real-Time PCR System (Agilent Technologies, USA). Cycle threshold (Ct) values below 32 were considered positive according to the manufacturer's recommendations.

For the purposes of this study, *Blastocystis* spp. and *D. fragilis* were classified as potentially pathogenic intestinal organisms, reflecting their uncertain but potentially clinically relevant pathogenicity.

Statistical analysis. Statistical analyses were performed using R software (version 4.5.3). Continuous variables were summarized as medians with interquartile ranges (IQRs), while categorical variables were presented as counts and percentages. Group comparisons were performed using the chi-square test or Fisher's exact test for categorical variables and the Wilcoxon rank-sum test for continuous variables, as appropriate. Factors associated with the presence of intestinal organisms were evaluated using logistic regression analysis, including age, gender, place of residence, and gastrointestinal symptoms as explanatory variables. A two-sided p-value <0.05 was considered statistically significant. No adjustment for multiple comparisons was applied.

Ethical approval. The study protocol was approved by the Bioethics Committee of the Medical University of Białystok, Poland (Approval No. APK.002.254.2025).

RESULTS

Study population. A total of 118 adults were included in the study. Median age – 39 years (Q1–Q3: 33–55 years). Females accounted for 66% of the study population, while males constituted 34%. Most participants resided in urban areas (74%), whereas 26% lived in rural settings. Gastrointestinal symptoms were reported by 84% of participants. The most frequently reported symptoms included abdominal pain (n = 55), diarrhea (n = 44), constipation (n = 41), abdominal cramps (n = 32), and nausea (n = 23).

Prevalence of intestinal organisms. Intestinal organisms were detected in 20 individuals (16.9%), while 19 individuals (16.1%) harboured pathogenic or potentially pathogenic parasites. The most frequently identified organism was *Blastocystis* spp. (n = 13), followed by *D. fragilis* (n = 5). Single cases of *G. intestinalis*, *E. vermicularis*, *E. coli*, and *E.nana* were also observed. Co-infections were identified in two participants. One participant had a mixed infection with *E. vermicularis* and *Blastocystis* spp., while another had a mixed infection with *D. fragilis* and *E. nana*. Both participants reported gastrointestinal symptoms. Detailed results are presented in Table 1.

Table 1. Prevalence of intestinal parasites detected in the study population

	n	%
Participants with any intestinal organism detected	20	16.9
Participants with pathogenic and potenthiallly pathogenic parasites	19	16.1
Enterobius vermicularis	1	0.8
Giardia intestinalis	1	0.8
Blastocystis spp.	13	11.0
Dientamoeba fragilis	5	4.2
Entamoeba coli	1	0.8
Endolimax nana	1	0.8

Both cases of infection with strictly pathogenic intestinal parasites (*E. vermicularis* and *G. intestinalis*) occurred in individuals residing in rural areas.

Association analyses. Statistical analysis revealed no significant association between parasite occurrence and gender, place of residence, or presence of clinical symptoms. The only factor significantly associated with intestinal parasite infection was age; infected individuals were significantly older than uninfected participants (p = 0.016) (Tab. 2, Fig. 2).

Table 2. Factors associated with intestinal parasite infection in the study population

Characteristic	No n = 991	Yes n = 191	p-value2
Age	38 (33–49)	60 (36–62)	0.016
Sex			0.4
Female	64 (65%)	14 (74%)	
Male	35 (35%)	5 (26%)	
Type of residence			>0.9
Urban	73 (74%)	14 (74%)	
Rural	26 (26%)	5 (26%)	
Symptoms			>0.9
N	16 (16%)	3 (16%)	
Y	83 (84%)	16 (84%)	

[1] Median (Q1-Q3); n (%)

[2] Wilcoxon rank sum test; Pearson's Chi-squared test; Fisher's exact test

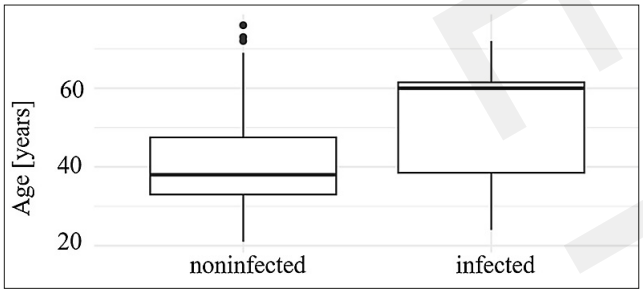

Figure 2. Age distribution according to intestinal parasite infection status

Age distribution among participants with and without intestinal parasite infection. Boxes represent the interquartile range (IQR), the central line indicates the median, whiskers extend to 1.5× IQR, and points represent outliers. Individuals with parasitic infection were significantly older than uninfected participants (p = 0.016, Wilcoxon rank-sum test).

DISCUSSION

This study demonstrated a relatively low carriage of intestinal parasites among adult residents of north-eastern Poland. These findings are consistent with observations from other high-sanitation European countries, where classical intestinal parasitic infections have become rare in the general adult population [1, 3, 11]. Nevertheless, the detection of intestinal organisms in nearly one-sixth of participants indicates that faecal–oral transmission has not been fully eliminated, even in highly developed settings.

A noteworthy finding was the predominance of *Blastocystis* spp. and *D. fragilis*, which together accounted for the vast majority of positive results. In contrast, unequivocally pathogenic parasites were detected only sporadically, with single cases of *G. intestinalis* and *E. vermicularis* identified in the studied population. This pattern is consistent with contemporary European epidemiological data, suggesting a shift from classical intestinal parasitic infections towards colonisation with uncertain pathogenicity [1, 2, 11].

Historical data from north-eastern Poland provide additional context for the present findings. In a study conducted among children and adolescents from the north-eastern region of Poland, Żukiewicz et al. reported intestinal parasitic infections in 75% of participants, with *A. lumbricoides*, *G. intestinalis*, and other classical intestinal parasites predominating [12]. In a long-term analysis of patients attending the Out-patient Division of Maritime and Tropical Medicine in Gdynia, northern Poland, Kowalewska et al. reported a decline in the prevalence of classical intestinal parasites over the period 1978–2010, accompanied by an increasing predominance of *Blastocystis* spp. [13]. More recent data from eastern Poland showed potentially pathogenic intestinal organisms in 9.2% of Border Guard officers from the Podlaski Border Guard Unit, with *Blastocystis* spp. accounting for the vast majority of positive findings [14].

Against this background, the 16.9% detection rate observed in the present study is within the range reported in previous Polish studies, although direct comparisons are limited by differences in study populations, diagnostic methods, and the spectrum of organisms investigated.

The clinical significance of *Blastocystis* spp. and *D. fragilis* remains controversial. While some studies associate these organisms with gastrointestinal symptoms and functional bowel disorders, others suggest that they may represent components of a diverse eukaryotic intestinal microbiota. Notably, *Blastocystis* spp. is currently considered a stramenopile rather than a classical parasite in a strict clinical sense, further complicating its interpretation in diagnostic and epidemiological contexts [6, 7, 15].

No significant association was observed between the presence of intestinal organisms and self-reported gastrointestinal symptoms. However, this finding should be interpreted with caution, as the majority of participants reported gastrointestinal complaints. Consequently, the study design does not allow exclusion of a potential role of organisms such as *Blastocystis* spp. or *D. fragilis* in symptom generation in individual cases. Rather, the results suggest that the mere presence of these organisms was insufficient to explain gastrointestinal symptomatology in the studied population. Gastrointestinal symptoms are common in the populations of high-income countries and may arise from

a broad spectrum of infectious, inflammatory, functional, metabolic, and microbiota-related conditions [16, 17]. Therefore, parasites of the gastrointestinal tract should be considered only one of many potential contributing factors to digestive complaints.

A strength of the study is the important observation of the significant association between age and the presence of intestinal organisms. Similar findings have been reported in other European populations, where colonisation with *Blastocystis* spp. and other intestinal protists is more frequent in older individuals [18, 19]. This association may reflect cumulative environmental exposure over the lifespan, as well as age-related changes in host immunity and intestinal microbiome ecology [1, 10]. However, the underlying mechanisms remain insufficiently understood and require further investigation.

No significant associations were found between parasite detection and gender or place of residence. Nevertheless, the relatively small number of positive cases may have limited the statistical power to detect weaker epidemiological relationships.

An additional strength of this study was the use of molecular methods targeting selected clinically relevant intestinal parasites. The absence of molecular evidence for most pathogenic species supports the conclusion that clinically significant intestinal parasitic infections are currently rare among adults in this region of Poland.

Limitations of the study. The study has several limitations. First, the sample size was relatively small, which limited the generalisability of the findings. Second, molecular testing did not include *Blastocystis* spp. or *D. fragilis*, preventing direct comparison between microscopic and molecular detection rates for the most frequently identified organisms. Additionally, information on place of residence was limited to an urban/rural classification and did not capture potentially relevant environmental and behavioural exposures, such as contact with animals, occupational exposures, food consumption practices, or household conditions. Therefore, the absence of an association with place of residence should be interpreted cautiously.

In summary, intestinal parasites and protists are still present in the adult population of north-eastern Poland, although classical pathogenic infections appear to be rare. The predominance of *Blastocystis* spp. and *D. fragilis*, together with the observed age-related association, suggests that intestinal eukaryotes remain an underappreciated component of adult gut ecology and epidemiology.

CONCLUSIONS

Intestinal parasites are still detected among adults in north-eastern Poland, although classical pathogenic infections appear to be uncommon. Among the organisms identified, *Blastocystis* spp. and *D. fragilis* predominated, while *G. intestinalis* and *E. vermicularis* were observed only sporadically. No association was found between parasite detection and gastrointestinal symptoms, whereas older age was significantly associated with the presence of intestinal organisms. Further studies involving larger adult populations and comprehensive molecular diagnostics are needed to better characterise the epidemiology and clinical relevance of intestinal protists in developed countries.

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