

Disruption of The Intestinal Microbiota in Parasitic Diseases in Children

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Abstract

This study investigated the prevalence of parasitic infections, clinical manifestations, and the state of intestinal microbiota in children under 5 years of age. The main complaints included abdominal pain, decreased appetite, sleep disturbances, perianal itching, allergic skin manifestations, and gastrointestinal and neurobehavioral symptoms. Coproscopic analysis revealed parasitic infections in 73.0% of the examined children. The most common forms were mono-enterobiasis (32.0%) and mixed infection of enterobiasis with giardiasis (23.0%).

Microbiota analysis demonstrated dysbiotic changes associated with parasitic invasions. A decrease in beneficial bacteria (*Bifidobacterium* and *Lactobacillus*) and an increase in opportunistic microorganisms (*Enterobacteriaceae*, *Escherichia coli*, *Klebsiella* spp.) were observed. These alterations were most pronounced in mixed infections.

The findings confirm that parasitic infections negatively affect intestinal microbiocenosis, leading to dysbiosis, which plays an important role in disease severity and clinical course. Therefore, a comprehensive treatment approach, including both antiparasitic therapy and microbiota correction, is essential.

Keywords: Parasitic infections, enterobiasis, giardiasis, intestinal microbiota, dysbiosis, children, helminthiasis.

Introduction

The gut microbiota plays a crucial bidirectional role in the course of helminth infections and is considered a key regulator of host susceptibility to parasites, parasite colonization in the intestine, and the development of immune responses. Helminth infections significantly alter the composition and diversity of the gut microbiota. These changes may either exacerbate pathological processes in the host or, in some cases, alleviate them [1; 3].

This tripartite interaction between the host organism, the microbiota, and helminths is of great importance for understanding the evolution of chronic inflammatory diseases and immune responses [5].

Key aspects of microbiota–helminth interaction include

the following: helminths shape the microbiota, meaning that helminth infection in many cases increases the diversity and richness of the gut microbiome. In particular, chronic helminth infections lead to an increased proportion of bacteria belonging to the *Lactobacillaceae* family in the intestine [3; 6].

Certain bacteria are essential for parasite colonization in the host organism. For example, the hatching of *Trichuris muris* (whipworm) eggs depends on signals derived from the gut microbiota. *Heligmosomoides polygyrus* is unable to efficiently establish a chronic infection in germ-free animals [2].

The microbiota altered under the influence of helminths promotes the expansion of regulatory T cells (Tregs) in the

intestine. These cells exert an immunosuppressive effect on immune responses. As a result, long-term parasite survival is facilitated, while at the same time allergic inflammatory reactions (e.g., bronchial asthma) are reduced [4].

Helminth infection increases the levels of metabolites produced by the microbiota, particularly short-chain fatty acids. These metabolites play an important role in regulating anti-inflammatory responses within the host immune system [7].

In industrialized countries, the elimination of helminths through antihelminthic therapy may lead to the loss of microbiota-mediated immune regulation. This condition is thought to contribute to an increased prevalence of autoimmune and chronic inflammatory diseases [8].

In conclusion, the interaction between the gut microbiota and helminths is a complex, dynamic, and bidirectional system that plays a crucial role in maintaining immune homeostasis. Disruption of this ecosystem balance is considered an important factor in the development of chronic inflammatory and immune-mediated diseases.

Accordingly, the aim of the study was to investigate the gut microbiota in patients with intestinal helminth infections.

Methods

A cross-sectional observational study was conducted in 100 children under the age of 5 years. All children underwent coprological examination. Stool samples were collected using formalin-containing PARASEP containers. This method is considered one of the most modern and highly sensitive techniques. Formalin fixes parasites, while ether dissolves fats. The purpose of the method is to detect hard-to-find eggs and protozoan cysts (e.g., *Giardia* and dysentery amoebae) by concentrating them in the sediment.

Approximately 1–2 grams of stool sample were thoroughly mixed in a test tube with 5–7 mL of 10% formalin solution. Formalin serves to preserve parasite morphology through fixation without structural damage. The resulting mixture is filtered through gauze or a special filter (e.g., the Parasep system) into another test tube to remove large food particles.

Then 2–3 mL of ether (or ethyl acetate) is added to the filtrate. The tube is tightly closed and vigorously shaken for 30 seconds. Ether dissolves fats present in the stool and facilitates the separation of debris. The tube is centrifuged at high speed (usually 2000–3000 rpm) for 2–3 minutes. After centrifugation, four layers are formed: the top ether layer, the second layer containing fat and debris, the third layer containing formalin solution, and the sediment at the bottom, where the parasites accumulate. The upper liquid layers are discarded. A smear is prepared from the sediment at the bottom and carefully examined under a microscope.

To assess the state of the intestinal microbiome, stool samples were collected in sterile containers and delivered to the bacteriological laboratory within 6 hours. The stool

samples were cultured using the classical bacteriological method, and the grown colonies (*Bifidobacteria*, *Lactobacilli*, *E. coli*) were quantified as CFU/g (colony-forming units per gram). The obtained results were subjected to statistical analysis.

Results and Discussion

The main complaints in children included intermittent abdominal pain, reduced appetite, insomnia, nocturnal teeth grinding (bruxism), perianal itching, nail-biting habit, irritability, skin discoloration, allergic skin rashes, constipation, diarrhea, enuresis, and halitosis upon waking in the morning. Based on coproscopic examination, the following diagnoses were established: *Hymenolepis nana* in 12 children (12.0%); enterobiasis combined with giardiasis (*Giardia lamblia*) in 23 children (23.0%); ascariasis in 6 children (6.0%), while no intestinal helminth infections were detected in 27 children (27.0%). The results showed that parasitic infections were present in 73.0% of the examined children. The most common findings were mono-enterobiasis (32.0%) and mixed enterobiasis + giardiasis infection (23.0%). Intestinal microbiota was assessed in all children. According to the results, bifidobacteria, the main representatives of normal intestinal microflora, were detected at the highest level in the control group ($10^{9.5}$).

In groups with parasitic diseases, a stepwise decrease in their levels was observed: in enterobiasis $10^{9.0}$, in ascariasis $10^{8.7}$, in hymenolepiasis $10^{8.8}$, and in mixed invasion $10^{8.0}$. A similar trend was also noted for lactobacilli: the level of $10^{7.5}$ in the control group decreased to $10^{7.2}$ in enterobiasis, to $10^{7.0}$ in ascariasis and hymenolepiasis, and further to $10^{6.8}$ in mixed invasion. This pattern indicates a reduction of beneficial microflora in the presence of parasitic infections. Among opportunistic microorganisms, bacteria belonging to the family Enterobacteriaceae accounted for $10^{6.5}$ in the control group, while their levels increased in parasitic diseases: $10^{7.2}$ in enterobiasis and ascariasis, $10^{7.5}$ in hymenolepiasis, and up to $10^{7.8}$ in mixed invasion. A similar pattern was observed for *Escherichia coli* (total), which was $10^{6.0}$ in the control group, increased to $10^{7.0}$ in enterobiasis and ascariasis, to $10^{6.5}$ in hymenolepiasis, and sharply increased to $10^{8.5}$ in mixed invasion. Enterococcus levels were relatively stable, amounting to $10^{5.5}$ in both the control and enterobiasis groups, but increased to $10^{6.5}$ in hymenolepiasis. *Klebsiella* spp. remained at $10^{4.5}$ in the control and enterobiasis groups, while rising to $10^{6.0}$ in ascariasis and mixed invasion (Table 1).

The study results demonstrated a high prevalence of parasitic infections in children (73.0%), which is of significant epidemiological importance. The most common conditions were mono-enterobiasis and mixed enterobiasis + giardiasis infection, confirming the predominance of contact–household transmission in pediatric populations. These findings are consistent with data reported in many studies, which indicate a high prevalence of parasitic diseases in children, often occurring in mixed infection forms.

Clinically observed symptoms—intermittent abdominal pain, decreased appetite, perianal itching, bruxism, insomnia, allergic skin rashes, and dyspeptic disorders—

are characteristic of parasitic invasions and can be explained by the mechanical, toxic, and allergic effects of parasites. In particular, perianal itching and teeth grinding (bruxism) are considered pathognomonic for enterobiasis.

Intestinal microbiota analysis clearly demonstrated the development of dysbiotic changes in the context of parasitic infections. The main representatives of normal microflora—bifidobacteria and lactobacilli—were reduced in all parasitic groups. This finding is widely supported in the literature and can be explained by the ability of parasites to alter the intestinal environment, damage the intestinal mucosal barrier, and modulate the immune system, thereby inhibiting the growth of beneficial microbiota.

According to modern studies, the interaction between the intestinal microbiota and parasites is bidirectional and operates within the “host–microbiota–parasite” system. Excretory–secretory products released by helminths, as well as extracellular vesicles, directly influence the composition of the microbiota. At the same time, they modify the production of antimicrobial peptides, thereby disrupting the balance of the microbial community.

In our study, an increase in the abundance of opportunistic pathogenic flora—Enterobacteriaceae, *Escherichia coli*, and *Klebsiella* spp.—was observed. These indicators were particularly elevated in mixed invasion cases. This condition is associated with competition for nutrients by parasites, alterations in the intestinal environment, and suppression of immune surveillance. According to the literature, parasitic infections aggravate intestinal dysbiosis and create favorable conditions for the overgrowth of opportunistic microorganisms. It has also been reported that helminth infections may increase intestinal microbiota diversity. However, this increase in diversity is not always functionally beneficial, as it may also result from the expansion of pathogenic or opportunistic bacterial populations. In our findings, a similar pattern was observed, characterized by a decrease in beneficial microbiota and an increase in opportunistic bacterial flora.

From an immunological perspective, parasites activate a Th2-type immune response while simultaneously suppressing immune activity through regulatory T cells (Tregs). This mechanism facilitates prolonged persistence of parasites in the host organism and contributes to the development of microbial imbalance within the gut microbiota. Alterations in the production of short-chain fatty acids (SCFAs) also influence inflammatory processes. In the context of mixed infections, the deeper progression of dysbiosis can be explained by the simultaneous action of multiple pathogenic factors. This condition may lead to the exacerbation of clinical symptoms and a reduction in treatment efficacy.

Thus, the obtained results confirm a complex interaction between parasitic infections and the intestinal microbiota. Parasites not only exert a direct pathogenic effect but also play an important role in disease pathogenesis by altering the intestinal microbiocenosis. Therefore, in the treatment of parasitic diseases in children, it is advisable not to limit therapy to anthelmintic treatment alone, but to apply a

comprehensive approach aimed at restoring the intestinal microbiota.

Conclusion

Monoinfections (enterobiasis, hymenolepiasis) lead to grade I disruption of the intestinal microbiome, whereas ascariasis and mixed infections result in more profound alterations corresponding to grade II intestinal microbiome dysbiosis.

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Table 1**Results of bacteriological examination of stool samples in the study group of children**

	Without parasitosis (n=27)	Enterobiasis (n=32)	Ascariasis (n=6)	Hymenolep iasis (n=12)	Mixed invasion (n=23)
Bifidobacterium	$10^{9.5}$	$10^{9.0}$	$10^{8.7}$	$10^{8.8}$	$10^{8.0}$
Lactobacillus	$10^{7.5}$	$10^{7.2}$	$10^{7.0}$	$10^{7.0}$	$10^{6.8}$
Enterobacteriaceae	$10^{6.5}$	$10^{7.2}$	$10^{7.2}$	$10^{7.5}$	$10^{7.8}$
E. coli (общая)	$10^{6.0}$	$10^{7.0}$	$10^{7.0}$	$10^{6.5}$	$10^{8.5}$
Enterococcus	$10^{5.5}$	$10^{5.5}$		$10^{6.5}$	$10^{5.5}$
Klebsiella spp.	$10^{4.5}$	$10^{4.5}$	$10^{6.0}$	$10^{4.5}$	$10^{6.0}$