Conceptions in parasite-microbiota relationships

Enas A Elsaftawy1,2, Rita M Wassef3

Departments of Medical Parasitology, Faculty of Medicine, Cairo University1, Armed Forces College of Medicine2, Helwan University3, Cairo, Egypt

ABSTRACT

Abundant information refers to the role of microbiota in maintaining homeostasis by assisting, improving, and regulating immunity in humans. Yet, the effect exerted by parasitic infections on the developing microbiota, is so far not recognized. Various parasites were related to specific alterations in the essential loads of developing microbiota. Additionally, modulation of immune functions was reported. This review aimed to present examples for the interaction between parasitic infections and microbial gut imbalance dubbed dysbiosis. This form of parasite-microbiota interaction seems to influence both the metabolism and the acquired immunity of the host. Consideration of this data will direct future studies to focus on the altered microbiota during various parasitic infections and to highlight the benefits of its re-enrichment in the therapeutic trials.

Keywords: dysbiosis; flora; luminal parasites; microbiota; probiotics

INTRODUCTION

Parasites are organisms that depend on their hosts, and typically in a parasitic relationship, one organism flourishes and proliferates at the expense of the other. Research on the normal microbiome communities during parasitic infections showed that the definition of ‘intestinal parasitosis’ requires re-assessment. It was postulated that intestinal dysbiosis as a state of disease induced by a parasite, is distinctive from the term intestinal parasitosis1,2.

The word ‘microbiota’ is composed of two syllables; ‘micro’ that refers to the invisible size, and ‘biota’ which is the bacterial ecosystem that inhabits a specific site in the host. This microbiota plays a crucial role in early life to establish the immune system in health and disease3,4. During early life, a stable gut microbiome is completed by the age of 31 months5,6, and its balance continues to be crucial for the long-term well-being and health7. Elwakil8 suggested that intimate factors and pathways governing the relationships between host, microbiota, and parasites are multifaceted. Although in several cases, the relation is not fully defined, a recent review has supported the role of microbiota in remedying the host immature immune system9.

Dysbiosis due to bacterial destabilization has been associated with several diseases. For instance, recent studies revealed alterations in intestinal bowel diseases10-12. Additionally, a recent study conducted by Ramírez-Carrillo et al.13 related the incidence of depression to the disturbance in the networks of the gut microbiota by Ascaris lumbricoides. However, modulation of this microbiota through consumption of live bacteria (probiotics), or non-digestible elements has been speculated in the inhibition or even treatment of some conditions14. The current review aimed to characterize some figures of alternations in microbiota associated with various parasitic infections.

[I] Intracellular parasites

1. Toxoplasma gondii is an intracellular sporozoan that can cause infection in any nucleated cell. Toxoplasmosis has been shown to induce massive necrosis and extensive pathology in the distal intestinal mucosa and villi in murine models. These pathological changes were attributed to bacterial dysbiosis besides immune factors such as the CD-4 T cells and interferon gamma (IFN-γ) cytokine15. Dysbiosis had been proposed to be dependent on the destabilization of the intestinal barrier either by the lytic replication of the parasite or from the toxic by-products of the host immune reactions, such as nitrogen intermediates or reactive oxygen species. For instance, production of IFN-γ during toxoplasmosis leads to loss of Paneth cells resulting in reduced levels of the antimicrobial peptides and dysbiosis16,17 dominated chiefly by Enterobacteriaceae18. In fact, the induced IFN-γ signal transducer and activator of transcription-1/inducible nitric oxide synthase (IFN-γ/STAT-1/iNOS) axis, was described as crucial for parasite resistance19. It was assumed to provide a pool of nitrate that acts as a source for anaerobic respiration and promotes overgrowth of Enterobacteriaceae.

Similarly, in experimentally induced colitis using dextran sulphate sodium (DSS), host-derived nitrate was found to directly promote the flush of Enterobacteriaceae bacteria in the colon20. This proves that local inflammation in the large intestine causes upgrade of the respiratory electron acceptors
levels like N oxides and S oxides, that act as a substrate for the expansion of facultative anaerobic bacteria\cite{29}, Hatter et al.\cite{30} noted an ominous outgrowth of Clostridia spp. that was sustained in chronic infection with toxoplasmosis. Also, Egan et al.\cite{31} suggested that the depletion of gut flora reduces mice resistance to T. gondii-triggered ileitis. Likewise, Couturier-Maillard et al.\cite{32} suggested that the collaboration between intestinal commensals and T. gondii in the development of small intestine inflammation increases the capability of the parasite to approach the epithelial cells and promotes its invasion to the mucosa.

**Cryptosporidium parvum** is another intracellular intestinal sporozoan that has been hypothesized to trigger dysbiosis indirectly by causing damage to the intestinal epithelium\cite{33}. Fecal microbiota from mice infected with *C. parvum* were shown to differ significantly from that of non-infected controls. Amazingly, a recent study demonstrated that probiotics can significantly influence the intestinal microenvironment and the epithelial lining which promotes the proliferation of *C. parvum*\cite{34}.

**Leishmaniasis** is caused by intracellular flagellates that were shown to trigger skin dysbiosis in humans and murine models. Cutaneous leishmaniasis is characterized by augmented *Staphylococcus* and/or *Streptococcus* infections and skin inflammation. Interestingly, in murine models this dysbiosis was found to be transferable to naive mice\cite{35}. Similarly, visceral leishmaniasis (VL) patients are more susceptible to fatal nosocomial infections\cite{36}. This was attributed to the suppressed status of T-helper 1 due to alterations in antigen presentation, MHC/HLA, antigen processing, T cell receptor recognition, and the myeloid derived suppressor cells\cite{37,38,39}. However, establishment of VL infection in murine models with previously induced intestinal dysbiosis showed late onset and development of weight loss\cite{40}.

**Blastocystis spp.** are the most common eukaryotic intestinal protozoa in humans\cite{41}. As an anaerobe, it lacks several definitive mitochondrial features and misses the standard mitochondrial electron transport chain and oxidative phosphorylation\cite{42}. Notably in a healthy gut, oxygen concentration is extremely low to promote the growth of obligate anaerobic microbiota from the Bacteroides and Firmicutes phyla\cite{43}. In a dysbiotic gut, when the intestinal microbiota get disrupted and luminal bioavailability of oxygen increases, the resulting alteration in intestinal biodiversity promotes the growth of the facultative anaerobic *Enterobacteriaceae*\cite{44,45,46,47}. A strict anaerobe such as *Blastocystis* spp. cannot sustain itself in this medium that is not its ideal ecosystem. In this context, in a well-established irritable bowel syndrome (IBS), dysbiosis *Blastocystis* spp. may be compelled out of the gut since the early stages of the disease\cite{48}.

On the other hand, Tsaousis et al.\cite{49} suggested that *Blastocystis* spp. has an alternative oxidase nature that allows it to deal with fluctuating oxygen concentrations in the gut and should be better termed as a microaerophilic. *Blastocystis* infection in rats involved colon hypersensitivity and showed alterations in the composition of the microbiota and hence their metabolic shifts\cite{50}.

Conversely, two studies\cite{51,52} reported the presence of healthy microbiota rather than dysbiosis in patients infected with *Blastocystis* spp. Moreover, Scanlan et al.\cite{53} considered *Blastocystis* spp. a member of the
healthy microbiota. However, in a previous study the genotype of the parasite appeared to play a hidden role in the capability of the parasite in contributing to disease\textsuperscript{[54]}. In this context, the relationship between different genotypes of the parasite and dysbiosis seems to be a new point of research.

(3) \textit{Entamoeba histolytica} was previously shown to modify the tight junction proteins and disturb the permeability of the epithelial barrier, allowing translocation of intestinal microbiota into the mucosal surfaces and spread to the other organs\textsuperscript{[55,56], 61}. Iyer et al.\textsuperscript{[57]} reported that \textit{E. histolytica} favours the phagocytosis of bacterial species such as \textit{Lactobacillus ruminus}. Similarly, Verma et al.\textsuperscript{[58]} revealed that \textit{E. histolytica} associated dysbiosis was characterized by increased \textit{Bifidobacterium} spp. in faecal samples, while other bacteria that are essential for the maintenance of intestinal homeostasis such as \textit{Bacteroides}, \textit{Clostridia}, \textit{Campylobacter}, \textit{Lactobacillus}, and \textit{Eubacterium} were reduced as compared to healthy subjects. Interestingly, Sicard et al.\textsuperscript{[59]} demonstrated that colonic microbiota break down complex carbohydrates into glycans that can act as a nutrient source for \textit{E. histolytica}. With the absence of bacteria, \textit{E. histolytica} is forced to be in close contact with the epithelium to feed and search for alternative sources of energy in the lumen\textsuperscript{[60]}. Moreover, in germ free mice the MUC2 layer was found to be thinner and penetrable\textsuperscript{[59]} with diminutive O-glycan monomers\textsuperscript{[54]).

Additionally, it was postulated that the ratio of bacterial phyla present in the host microbiota may alter or influence the colonization of \textit{E. histolytica}. For instance, \textit{Escherichia coli} was found to protect \textit{E. histolytica} against oxidative stress in two patterns\textsuperscript{[61]} described in figure (1). However, in murine models \textit{Clostridium} is defensive during \textit{E. histolytica} infection through the increased production of interleukin 17-A, dendritic cells, and neutrophils. Moreover, bone marrow-derived dendritic cells were reported to secrete elevated levels of IL-23\textsuperscript{[62]}. Gilchrist et al.\textsuperscript{[63]} showed that the expansion of \textit{Prevotella copri} probiotic was associated with dysentery due to intestinal amoebiasis in infants from 600 to 800 days of age. Sarjapuram et al.\textsuperscript{[64]} revealed that probiotics (specifically \textit{Lactobacillus casei}) have an anti-proliferative effect on \textit{E. histolytica}. Varet et al.\textsuperscript{[61]} speculated this effect to be unrelated to alterations in the pH value of the media.

(4) \textit{Trichuris trichiura} is a round worm reported in a Malaysian study to be associated with shifts in the composition of the gut microbiota of infected patients especially with \textit{Paraprevotellaceae} bacteria concerned with the breakdown of proteins and carbohydrates in food\textsuperscript{[65]}. In general, helminthic infections may promote regulatory immune cell reactions that may suppress harmful allergic and inflammatory diseases. They are supposed to elicit regulatory T cells by altering the gut microbiota or via secretion of bioactive molecules. Hence, worms and their secretory molecules may be innovative treatments for allergic inflammation\textsuperscript{[1,66]}

(5) \textit{Trichomonas vaginalis} is a flagellate that causes the most common sexually transmitted disease (STD) during the child bearing period in female patients\textsuperscript{[67]}. In fact, trichomoniasis was shown to pave the way for bacterial dysbiosis owing to multiple virulence factors. This protozoon employs its cytopathic effect through the release of pore-forming proteins active at pH 5.8\textsuperscript{[68]}. Garber et al.\textsuperscript{[69]} isolated a specific soluble heat and acid labile cell-detaching factor from cultured \textit{T. vaginalis} that possesses trypsin like action and can disrupt the monolayer cells at optimum pH 6.5. In another experimentally infected murine model, β-hemolysin was reported to be another virulence factor\textsuperscript{[69]} that destructs the carbohydrate cell monolayers and was found to produce subcutaneous abscesses\textsuperscript{[70]}. Invasion of the mucous layer entails efficient motility by the parasites and the production of adhesins and mucinases\textsuperscript{[71]}. Additionally, to thrive the antibody response, it produces several cysteine proteinases that degrade IgG, IgM, and IgA\textsuperscript{[72]).

Increased \textit{T. vaginalis} colonization was associated with bacteri al \textit{Gardennella vaginalis} species that cause bacterial vaginosis at the expense of the commensal \textit{Lactobacillus} spp.\textsuperscript{[73]). This was attributed to the efficient

---

**Escherichia coli** live bacteria protects \textit{E. histolytica} against oxidation by two patterns of mechanisms

<table>
<thead>
<tr>
<th>1. Reduction in the expression of genes encoding</th>
</tr>
</thead>
<tbody>
<tr>
<td>• Protein synthesis (ribosomal proteins and translation related factors).</td>
</tr>
<tr>
<td>• Proteins essential for parasite nutrition (hydrolases, peptidases, and trafficking).</td>
</tr>
<tr>
<td>• Members of LRR family.</td>
</tr>
<tr>
<td>• Anti-oxidant enzymes.</td>
</tr>
<tr>
<td>• Encystation factors.</td>
</tr>
</tbody>
</table>

| 2. Reverse in the expression of genes encoding for amoeba survival, e.g. genes for oxidoreductase enzyme. |

---

**Fig. 1.** Paradigm of \textit{Escherichia coli}/\textit{E. histolytica} interaction. LRR: Leucine-rich repeat family genes. Illustrated by E. Elsaftawy.
phagocytic action of the parasite by killing and ingesting lactobacilli[75]. Diminution in lactobacilli populations upgrades the vaginal pH that is normally fluctuating between 2.8 and 4.2[76] creating a milieu more promising for the proliferation of dysbiotic bacteria. Subsequently, the biofilm of the dysbiotic bacteria was found to provide adhesion for T. vaginalis[78]. Also, dysbiotic bacteria refine the cytoadhesion of the parasite to the vaginal cells and muncins. Gardnarella vaginalis is the most associating dysbiotic bacterium; however other genera, such as Atopobium vaginae, Prevotella bivia, Bacteroides, and Mycoplasma hominis can also flourish[67,73,78,79].

**[III] Blood flukes (Schistosoma spp.)**

*S. haematobium* is a urinary blood fluke (trematode) that inhabits the vesical plexus around the bladder. It has been related to a shift in the structure of the faecal microbiome community[80-82], in the form of reduced phylum Firmicutes bacteria and prevalence of phylum Proteobacteria bacteria. A number of alterations were also detected including reductions in Clostridia family bacteria and surges in Moraxellaceae, Veillonellaceae, Pasteurellaceae, and Desulfovibrionaceae[81]. Enhancement of urease enzyme has been related to dysbiosis and inflammation. Owing to the habitat of the parasite in the vasculatures around the urinary bladder the indirect link between systemic immunity and microbiota can be suggested as a point of research[81].

*S. mansoni* is another blood fluke that inhabits the inferior mesenteric plexus of veins draining the colon. Using murine models, a partial relationship was proposed between the parasitic infection and changes in the configuration of the host microbiome[83]. Variances in the gut microbial paradigm involved enrichment in Proteobacteria phylum and the deficiency of the immunoregulatory bacteria (e.g. Lactobacillus). This disruption in the gut microbiota was directly associated with higher worm and egg burden, together with amplified immune responses to Schistosoma antigens[84]. Interestingly, higher abundance of Fusobacterium spp. was found to be accompanied with increased efficacy of praziquantel[85]. On the other hand, Jenkins et al.[86] and Schneeberger et al.[87] proposed that neither S. mansoni infection nor praziquantel administration promotes a significant alteration in the composition of gut microbiota.

Another interesting finding was published by Floudas et al.[88] who reported less susceptibility to induced colitis in mice infected with adult male S. mansoni as compared to infection with adult male and female worms. This was assumed to be related to the capability of the male worms to regulate the host’s immune reactions and overcomes colitis through restricting gut dysbiosis.

**[IV] Skin parasites**

*Sarcoptes scabiei* is a mite that causes skin infestation and is thought to promote bacterial infections by breaching the skin barrier. It incapacitates the expression of many adhesion molecules of the epidermal cells, the endothelial cells and the dermal fibroblasts[87-89]. This is achieved by secretion of protease enzymes to facilitate tissue invasion and migration[90]. It evades immunity through the excretion of uncharactized immune modulatory molecules[91] and complement inhibitors that are supposed to assist the propagation of *Streptococcus pyogenes*[92]. Moreover, it induces itching due to allergy and inflammatory responses against the arthropod and its secretory/excretory metabolic by-products that pave the way for bacterial infections. Abreu-Velez et al.[93] assumed that itching sensation may be aggravated as an auto-reactivity to sweat glands and nerves in close proximity to the sites of infestation.

Disruption of the skin barrier facilitates the invasion of opportunistic pathogens and permits secondary bacterial infection, most frequently by *Staphylococcus aureus* and *S. pyogenes* that belongs to group A Streptococci (GAS). These bacteria have been isolated from skin tunnels and faecal by products of the mite suggesting that the arthropod could contribute directly to the dissemination of the bacteria and increase risk of impetigo[94-95]. Moreover, Swe et al.[96] using a porcine model, assumed that scabies mites shift the growth of the skin microbiota from the commensal *Staphylococcus hominis* to *Staphylococcus chromogenes*. Further sequel of skin dysbiota is the real possibility of renal glomerular damage due to streptococcal infection that may occur many years afterwards[97] (Figure 2). Another study showed that control of scabies with ivermectin is also related to significant lower isolations of streptococci from skin lesions and haematuria[98].

**CONCLUDING REMARKS**

1. It is currently accepted that the gut microbiota interrelate with human health and that its disruption by a parasite has a hidden effect in the progression of the parasitic disease.
2. The real effect of dysbiosis in many parasitic diseases is still not clear and necessitates further research.
3. The current recommendations for probiotic therapies in parasitic diseases are still limited and requires further large cohorts' studies to determine the right probiotic strain for a given parasitic strain.
4. Parasites alter microbiota through different mechanism, as proposed in the following parasites:
   - *T. gondii*: Destabilization of the intestinal barrier, and increased production of nitrates.
   - *Cryptosporidium spp*: Damage to the intestinal epithelium.
   - *Leishmania spp*: Alterations in antigen presentation, MHC/HLA, and antigen processing, disruption of the T cell receptor recognition, as well as modification in the myeloid derived suppressor cells.
   - *G. lamblia*: Disruption of the integrity of MUC2, reduction of the viscous physical barrier, and degradation of tight junction proteins and apoptosis.
of the intestinal epithelial cells utilizing secretory-excretory cysteine proteases.

- **Blastocystis spp.**: Colon hypersensitivity, alterations of microbiota composition, and metabolic shifts of the intestinal microbiota
- **E. histolytica**: Several mechanisms were proposed: modification of goblet cell, reduction of mucus assembly (germ free murine models), modification of tight junction proteins, distortion of epithelial barrier permeability, translocation of intestinal microbiota into mucosal surfaces, dissemination of microbiota to other organs, gene regulation and altered expression of the leucine-rich repeat (LRR) family genes (documented with enteric bacteria but not with probiotics), phagocytosis of beneficial bacteria, and increased *Bifidobacterium* species.
- **T. trichura**: Promotion of regulatory immune cell reactions, and secretion of bioactive molecules.
- **Schistosoma spp.**: Enhancement of urease enzyme, immunomodulation, and alterations of microbiota composition.
- **S. scabiei**: Breach skin barrier was proposed through skin scratching (auto reactivity), and protease enzymes. Besides, immunomodulation and inhibition of the adhesion molecules were reported.

**Authors’ contribution**: Both authors contributed substantially to the manuscript. Elsaftawy EA conceptualised the idea, wrote the initial draft, and drew all the figures. Wassef RM supported the idea, revised and edited the manuscript, and took correspondence responsibility. Both authors agreed on the final version submitted to the chosen journal and approved revision in response to reviewers’ comments.

**Declarations of interest**: The authors declared that there were no potential conflicts of interest concerning the research, authorship, and/or publication of this article.

**Financial support**: This work did not receive any financial fund from any organization or institution.

**REFERENCES**


Perturbation of the intestinal microbiota of mice infected

Ras R, Huynh K, Desoky E, Badawy A, Widmer G. Toxoplasma gondii and microbiota contribute to the exacerbation of


