Abstract: Mucosal immunity and trained innate immunity of the gut play a pivotal role in maintaining intestinal homeostasis and defending against microbial pathogens. This review provides an overview of the mechanisms underlying mucosal immunity and the concept of trained innate immunity in the gut. We discuss the interaction between gut microbiota and the host immune system, highlighting the role of epithelial cells, dendritic cells, and innate lymphoid cells, as well as the novel concept of trained innate immunity and its role in perpetuating or attenuating gut inflammation. We also comment on the current models for investigating mucosal immunity, their limitations, and how they can be overcome. Additionally, we explore the potential therapeutic implications of modulating mucosal immunity and trained innate immunity in gastrointestinal diseases. Only by elucidating the mechanisms underlying mucosal immunity and the concept of trained innate immunity, innovative approaches to modulate immune responses and restore intestinal homeostasis in the context of gastrointestinal disorders could be implemented.

Keywords: mucosal immunity; trained innate immunity; gut microbiota; intestinal homeostasis; gastrointestinal diseases

1. Introduction

The mucosal immune system is a tightly regulated mechanism that primarily aims to prevent inappropriate immune responses to food or other antigens and protect the mucosa from invading pathogens. Antigens can access the mucosal immune system through several immune processes [1]. Mucosal immune responses occur at inductive sites in the mucosa-containing lymphoid tissue. This tissue is called mucosa-associated lymphoid tissue (MALT). In most cases, these mucosal immune reactions are triggered by viruses that infect mucosal surfaces [2].

The human gut microbiota participates in multiple interactions throughout the life of a host with various pathogens that could seriously affect host health. However, one of the fundamental mechanisms for maintaining intestinal homeostasis is colonization of the intestine, which occurs as early as the early neonatal age. The establishment and interactive...
development of this early gut microbiota is thought to be regulated by specific compounds already present in human milk [3].

Various metagenomic studies have emerged in the last decade that aim to analyze the composition and functionality of the gut microbiome from infancy. This type of analysis has linked certain features of the microbiota/microbiome, such as reduced diversity or abnormal composition, to gut diseases that occur later in life. The results obtained were almost identical, and they indicate that the early composition/development of the intestinal gut microbiota may influence the risk factors associated with health conditions in adults [4,5].

Probiotics, which are live microorganisms that provide health benefits to the host when administered in sufficient amounts, are appealing candidates for stimulating trained immunity; however, despite numerous studies documenting their beneficial properties, their mechanisms of action are not fully understood. Cortes-Perez et al. proposed investigating the possible link between probiotics and the promotion of trained immunity [6].

In line with this, many diseases result from a complex interaction between host genetics, environmental factors, and microbial factors. The relationship between the gut microbiome and some diseases has been attributed to the constant activation of the host immune system by the gut microbiota. The result of this dysregulated interaction between the host epithelium and microbes is a state of chronic inflammation, which in a significant number of cases is followed by the development of autoimmune, precancerous, and neoplastic diseases [7].

According to literature data, numerous scientific studies conducted on animal models and humans show that the immune system is essential in protecting against potential pests and maintaining immune tolerance to self-antigens. Although B cells play a crucial role in autoimmune diseases, understanding the intricacies of their effector functions remains a challenge to this day [8,9].

The immune system is a complex concept that includes a collection of cells, mediators, and chemical processes that protect against foreign antigens, not only individual systems but also all systems in the human organism. Beyond the structural and chemical barriers that protect us from infection, the immune system has two separate but interconnected lines of defense: innate and adaptive immunity. Most often, such disorders in one or both lines of the immune defense lead to disorders that could be broadly classified into three groups: hypersensitivity reactions, autoimmune, and immunodeficiency [10,11]. Conversely, the concept of trained immunity is related to innate immune responses. It represents a functional state of innate immune cells, mainly due to long-term epigenetic reprogramming [12].

This novel concept arose in infectious diseases: training innate immune cells (monocytes, macrophages, and/or natural killer cells, NK) through infection or vaccination improves immune responses to microbial pathogens after restimulation [13]. Although initially reported in circulating monocytes and tissue macrophages (known as peripheral trained immunity), subsequent research has shown that immune progenitor cells in the bone marrow can also be trained (known as centrally trained immunity), explaining the long-term innate-immunity-mediated protective effects of vaccination against heterologous infections [13].

Although trained immunity is effective against pathogens, improper activation by endogenous triggers can cause abnormal inflammation. For example, trained immunity may add to inflammatory activity in systemic lupus erythematosus (SLE) and systemic sclerosis, which accelerates disease progression [14,15].

This review provides an overview of the mechanisms underlying mucosal immunity and the concept of trained innate immunity in the gut by discussing the interaction between gut microbiota and the host immune system and highlighting the role of epithelial cells, dendritic cells (DCs), and innate lymphoid cells, as well as the novel concept of trained innate immunity and its role in perpetuation or attenuation of the gut inflammation. Additionally, we explore the potential therapeutic implications of modulating mucosal immunity and trained innate immunity in gastrointestinal diseases.
2. Search Strategy

We comprehensively searched multiple databases, including PubMed/MEDLINE, Embase, Scopus, and Web of Science. We used a combination of MeSH terms and free-text keywords to identify the relevant literature. MeSH terms such as “Gastrointestinal tract”, “Immunity, mucosal”, and “Immunity, innate” were combined with free-text keywords including “trained immunity”, “intestinal immunity”, and “gut microbiota”.

We combined search terms using Boolean operators: ("trained innate immunity" OR "trained immunity") AND ("gut" OR "gastrointestinal" OR "intestinal") AND ("Mucosa" OR "Intestinal Mucosa" OR "Gastrointestinal Mucosa") AND ("mechanisms" OR "epigenetic changes") AND “tolerance” OR “inflammation”). This approach enabled us to narrow down our search and focus on articles specifically related to the intersection of trained innate immunity, mucosal immunity, and gastrointestinal diseases for a time period of January 1960 to May 2024. We choose these types of papers: original research articles, review articles, meta-analyses, clinical trials, and case studies.

Additionally, we applied filters for human studies and the English language. By systematically searching the literature using these criteria, we aimed to identify relevant articles providing insights into the role of trained innate immunity and mucosal immunity. Approximately 700 papers were initially retrieved and then screened for relevance based on title and abstract, resulting in 84 papers thoroughly reviewed for inclusion in the study.

3. Brief Overview of the Gastrointestinal (GI) Immune System

MALT forms a crucial part of the body’s immune organ, termed the gut-associated lymphoid tissue (GALT) system. Collectively, MALT forms the largest immune organ in the body, as highlighted by Wershil et al. [16]. Inductive and effector sites encompass the two main compartments of the intestinal immune system. Inductive sites are comprised of Peyer’s patches (PP), mesenteric lymph nodes (MLN), and isolated lymphoid follicles (ILF). They take part in the initial priming and differentiation of immune cells.

Conversely, effector sites such as intestinal lamina propria and epithelium aid in maintaining adaptive immune cells and barrier integrity [17]. GALT is further categorized into aggregated and non-aggregated lymphoid tissues. The aggregated components comprise intestinal PPs, the appendix, and abundant lymphoid nodules. In contrast, non-aggregated lymphoid tissue includes intraepithelial lymphocytes and lamina propria lymphoid cells [18,19].

The next step of mucosal immune response is transport of the activated antigen-presenting cells to the lymph nodes—where their maturation in the form of T and B cell responses occurs. The formed virus-specific T and B cells, in turn, enter the blood. Finally, they return to the lamina propria of mucosal tissues, where they act as effectors (i.e., IgA-producing B cells) or reside as memory cells [20]. Preluence sites for mucosal immune responses were initially described in mucosal sites such as PPs in the small intestines [21].

The mucosal immune response resulting from primary viral infection is rapid. It is detectable in the mucosa as early as the third day after the first infection [20,22]. At the same time, this response peaks in the first 6 weeks and begins to decline after the first three months. According to the literature, this short duration of the primary mucosal antibody response is the host’s susceptibility to the particular disease after re-infection. In addition, some authors attribute this susceptibility to re-infection with a significant decrease in the number of IgA-antibody-secreting cells in the mucosa [23,24]. This is because re-infection or antigen boosting leads to a secondary immune response, which is characterized by a significantly faster rise in the titer of IgA antibodies and the maintenance of their high levels for a longer time.

Mowat et al. [17] remarked that collectively, the various immune components of the gastrointestinal system serve an essential role in facilitating the preservation of barrier function in its role of digestion and nutrient uptake. This is further evidenced by underlining that the intestine contains the most abundant immune cells of any tissue in the body. The gastrointestinal mucosa includes numerous variations of immune cells: T cells, B cells, and
DCs, as well as unique cell types such as PPs, intraepithelial lymphocytes (IELs), microfold cells, Paneth cells, and intestinal epithelial cells [16]. IELs are found in the intestinal epithelium above the basement membrane—they form a heterogeneous group of lymphocytes that are mainly effector memory T cells [16]. Noteworthily, subsets of IELs possess the unique ability to express CD8 [25]. Rather interestingly, unlike other T-cell populations, their development is independent of antigen presentation. The density distribution of IELs varies widely with antigen exposure, age, and location along the intestine [16].

Moreover, PPs are significant: clusters of immune cells consisting of a predominant B-cell follicle, an interfollicular T-cell region, and various macrophages and DCs. PPs are a substantial site for IgA1 B-cell development, which occurs under the influence of signals from T cells, DCs, and cytokines [26]. These lymphoid structures are located beneath a single layer of columnar cells known as the follicle-associated epithelium (FAE) [16]. Fu et al. further noted that the development of PPs relies on various factors, such as the interleukin (IL)-7 receptor and members of the tumor necrosis factor (TNF) receptor family [27].

Furthermore, Mowat et al. illustrated the importance of the specialized microfold cells (M cells) found in the follicle-associated epithelium of PPs and isolated lymphoid follicles (IFs). M cells are responsible for the uptake and transfer of antigens from the lumen into the subepithelium, where they encounter DCs and are subsequently presented to the adaptive immune cells [17]. Sampling of luminal antigens and microorganisms is believed necessary for developing immune responses and tolerance, as emphasized by Wershil et al. [16].

Another specialized group of cells, Paneth cells, are a distinct type of epithelial cell that participates in innate immunity. They can be found in the intestinal tract and play a role in innate immunity. Situated at the base of Lieberkuhn’s crypts in the small intestine, these cells contain various antimicrobial peptides, including α-defensins, lysozyme, and secretory phospholipase A2. Studies have identified and investigated six α-defensins, showing specific activity against both Gram-positive and Gram-negative bacteria and potential effectiveness against viruses [27].

Despite the primary function of the intestinal epithelial cells being the absorption of nutrients, it has become evident that epithelial cells also play an active part in mucosal immune responses. Intestinal epithelial cells can act as nonprofessional antigen-presenting cells, recognizing and responding to bacterial and viral patterns and producing various cytokines and chemokines that can impact immune responses [28,29]. Also, mast cells are predominantly located in the lamina propria and submucosa, with a small number in the epithelium [30,31].

These cells perform a crucial function by producing mediators that regulate several physiological processes, such as epithelial barrier integrity, peristalsis, vascular tone, and permeability. Finally, eosinophils are responsible for essential effector functions during parasitic infection and allergic responses, but they also contribute to normal gut homeostasis [32].

Immunoglobulin A (IgA) is the predominant antibody type in GALT [33]. IgAs are categorized into two classes, IgA1 and IgA2, with IgA2 being the primary form, and it is found mainly as secretory immunoglobulin A (sIgA). They function by blocking the entry of luminal antigens, microorganisms, and foreign proteins into the intestinal surface. Additionally, they also can deactivate toxins and infectious organisms [34].

IgM also plays an essential role in GALT. Its serum form protects against pathogens and maintains homeostasis but is less stable than IgA. However, secretory IgM (sIgM) may increasingly contribute to sIgA production and partially compensate for its absence in individuals with selective IgA deficiency, as shown by Fadallah et al. At the same time, it binds a broader range of gut bacteria and subsets of commensals with a higher level of microbial coating [35]. Another important immunoglobulin class in the submucosa is IgG. In humans, gut inflammation triggers the production of IgG1 and IgG3, while a healthy gut promotes the secretion of IgG2 and IgG4. Unlike sIgA, serum IgG has distinct targets...
within the gut microbiota, indicating a unique protective function against certain, but not all, commensals [36].

Although primarily recognized as a B-cell antigen receptor, IgD also can be found as a secreted antibody. IgD is secreted by IgM−IgD+ mucosal B cells that have experienced IgM-to-IgD class switch recombination (CSR). Secreted IgD could potentially aid in removing typical environmental substances from the body, such as allergens. Interestingly, IgE may also perform a comparable role, suggesting potential collaboration between IgD and IgE [37].

4. Mucosal Immune Responses in the Gut and the Novel Concept of Trained Immunity

Unlike the innate immune response, which has no immunological memory and is, therefore, unable to recognize the same pathogen if the body is re-exposed to it, acquired or adaptive immunity is antigen-dependent and specific and, therefore, followed by a maximal immune response. To this end, there exists the ability of adaptive immunity for immune memory, which allows the host to mount a more rapid and effective immune response upon subsequent exposure to the present pathogen. These two types of immunity are by no means mutually exclusive mechanisms but rather complementary, with defects in either system leading to diseases of a different nature [10,38].

The immune paradigm of innate vs. adaptive immunity is based on several aspects that distinguish these forms of immunity, including immunological memory. It is assumed that immune memory is attributed to adaptive immunity alone. However, this concept is now challenged [12]. Katz et al. pointed out that non-vertebrates evolved their innate immunity to have primitive immune memory exerted through re-infection with parasites [39]. Subsequently, the innate immune memory was documented in other non-vertebrates and vertebrates, including humans, supporting the hypothesis that this phenomenon is ubiquitous and evolutionary conserved [40].

The immunological memory response that innate immune cells might develop in response to prior insults is referred to as “trained immunity”, and it was initially coined in 2011 [40]. Even while cells revert to an inactive state following an initial stimulus, epigenetic modifications facilitate a more potent and rapid response when antigen is reintroduced [41]. The typical inducers of this type of training are pathogen-associated molecular patterns (PAMPs) and damage-associated molecular patterns (DAMPs), which can produce trained immunity in the bone marrow’s HPSCs (central trained immunity) and the circulation and tissues (peripheral trained immunity) [42]. We present the development and mechanisms of trained innate immunity in Figure 1.

Importantly, aberrant innate immune responses may lead to autoimmunity and chronic inflammatory disorders, even as trained immunity has developed under constant evolutionary pressure to offer improved protection against pathogens [43].

Hajishengallis et al. discussed trained innate immune cells in the mucosa, focusing on inflammation [44]. Based on the accumulated data, trained immunity may also encourage inappropriate immune responses that worsen pathology in contemporary settings where chronic mucosal and systemic inflammatory disorders are highly prevalent. Therefore, depending on the situation, defined agonists could be used therapeutically to either suppress excessive inflammation in inflammatory and autoimmune diseases or promote innate immune responses (especially helpful for treating infections or myelosuppression brought on by chemotherapy). Furthermore, trained immunity in mammals has evolved to protect against mucosal and systemic infection [45].
Figure 1. Trained innate immunity development and mechanism. Exposure to microbial signals and cytokines (PAMPs, DAMPs, etc.) educates myelomonocytic cells to have better effector function against microbes. Training can occur at either the bone marrow hematopoietic stem cell or mature macrophage levels. Training-induced enhancement of innate cell activity is dependent on epigenetic modifications. Trained myeloid cells have a higher killing ability and produce more cytokines, chemokines, and fluid-phase pattern recognition molecules. Furthermore, they are better able to elicit adaptive immune responses. PAMPs: pathogen-associated molecular patterns; DAMPs: damage-associated molecular patterns; oxLDL: oxidized low-density lipoproteins; TLR: Toll-like receptor; BCG: Bacillus Calmette–Guérin; LPS: lipopolysaccharides; Mφ: macrophages. Parts of the figure were drawn by using pictures from Servier Medical Art. Servier Medical Art by Servier is licensed under a Creative Commons BY 4.0 (https://creativecommons.org/licenses/by/4.0/ Accessed on 30 July 2024) and [f1-geg-8-2016-043] from the National Institutes of Health, In the Open NIH database (available at https://openi.nlm.nih.gov/detailedresult?img=PMC5038610_geg-8-2016-043f1&query=epigenetic%20modifications&it=xg&req=4&npos=16 Accessed on 30 July 2024).

5. Mechanisms of Trained Immunity

Recent studies have shown that innate immune cells acquire a trained immunity state through metabolic, epigenetic, and transcriptional reprogramming that lasts for months [46–51]. Trained immunity entails epigenetic and metabolic reprogramming of innate immune cells, resulting in qualitatively and quantitatively modified responses to later time-delayed heterologous activation. Misguided immune reactions can lead to disease progression, causing chronic hyperinflammation or immunological tolerance. Immunological tolerance dampens the host’s inflammatory response to maintain homeostasis and prevent tissue damage and organ failure but also increases the risk of secondary infections and other immune-related diseases [40].

The underlying processes of nonspecific enhanced responsiveness in innate immunity cells are based on epigenetic, transcriptional, and metabolic programming following transitory stimulation [52]. Changes to these programs result in improved reactivity to secondary challenges involving various stimuli. This behavior is called “trained immunity” or “innate immune memory”. On the one hand, trained immunity increases the response to infections and vaccinations, allowing for more excellent innate immune responses and better protection against a wide range of microbial stimuli. On the other hand, trained immunity may play a role in the pathogenesis of cardiovascular, autoinflammatory, and
neurodegenerative illnesses [52]. Netea et al. defined trained immunity as a biological process, exploring the innate cues and the epigenetic and metabolic reprogramming activities that influence trained immunity induction [40].

The idea of trained immunity refers to the long-term functional reprogramming of innate immune cells induced by exogenous or endogenous shocks. This results in an altered response to a subsequent challenge after returning to a non-activated state. The secondary response to the subsequent nonspecific stimulus can be modulated so that the cells respond more or less strongly than the primary response, resulting in context- and time-adjusted responses [41]. Trained immunity refers to the long-term adaptation of innate immune cells, not a specific transcriptional or functional program. Different stimuli, such as β-glucan, LPS, or the BCG vaccine, can trigger different trained immunity programs [41]. Furthermore, knowledge about the characteristics of trained immunity will help us comprehend host defense systems and the etiology of immune-mediated illnesses [40].

The immunological phenotype of trained immunity has been shown to endure at least 3 months and up to a year. In contrast, heterologous protection against illnesses caused by live vaccines can last up to 5 years [53]. Even so, trained immunity is often reversible and less long-lasting than traditional epitope-specific adaptive immunological memory [54,55]. Importantly, recent research has demonstrated transgenerational effects by creating trained immunity [56,57].

The main characteristics, mechanisms, and associated disorders with trained innate immunity are presented in Figure 2.

Figure 2. The concept of trained innate immunity: changes in the cells after the first stimulus, leading to trained immunity for the second stimulus by enhanced and improved immune response.

6. Disorders Affecting Gastrointestinal Immunity with a Focus on Mucosal Immunity

6.1. Inflammatory Bowel Disease (IBD)

The chronic gastrointestinal inflammation combined with the disturbed intestinal microbiota is the hallmark for diagnosing IBD—a group of associated diseases containing Crohn’s disease (CD) and ulcerative colitis (UC). In the pathogenesis of IBD, an immunological imbalance is implicated where the adaptive immune system cells become activated by self-antigens, leading to chronic inflammation [58].

As Lu et al. illustrated, microbiota dysbiosis contributes to this inflammatory state by promoting the growth of harmful microorganisms while allowing the loss of the beneficial ones. The result is impaired immune homeostasis with activation of the innate immune system and disturbance of the mucosal integrity [59]. Gut tissue damage is initiated and
sustained through excess production of pro-inflammatory cytokines, notably IL-1β, TNF-α, IL-18, and IL-17. The latter, released by Th17, activates the signal transducer and activator of transcription 3 (STAT3), which continues the vicious cycle of chronic inflammation by potentially promoting further Th17 differentiation and proliferation [59].

As Lee et al. noted [60], UC severity strongly correlates with the amount of IL-17 expressed by mononuclear cells in peripheral blood. The CD is most commonly associated with a Th1/Th17 inflammatory response, whereas atypical Th2 responses and Th17 inflammation predominate in ulcerative colitis. Following these facts, it is no surprise that the ratio Th17/Treg has an important role. When Tregs exceed Th17 levels, an anti-inflammatory state is promoted, whereas an abundance of Th17 has the opposite effect. Mutations in genes controlling Treg differentiation, e.g., CD25 and IL-10, have increased IBD occurrence. Loss of the anti-inflammatory effect of IL-10 also tips the scale towards inflammation. Additionally, innate lymphoid cells in the mucosa interact with mucosal cells and Tregs and respond accordingly with the secretion of IL-17, IL-22, and IFN-γ, contributing to the pool of pro-inflammatory cytokines [60].

Regarding trained innate immunity and IBD, the connection between them is not direct. Few studies explored the epigenetic impact of bacteria (i.e., lactic acid bacteria (LAB)) on the inflammatory pathways, including IL-17 and IL-23 expression in LPS-induced IBD models. The authors showed reduced histone acetylation and NF-kB activity and increased DNA methylation under the influence of two probiotic bacteria (Bifidobacterium breve and Lactobacillus rhamnosus GG) [61]. This model, although broadly used, has some limitations, such as germ-free laboratory environment and restricted microbiome composition. The limitations of the current animal models will be summarized in the next section of the review.

6.2. Infectious Diseases and Mucosal Immunity

Infectious diseases of the GIT can reveal a virus, a bacterium, or a parasite as the pathogenic microorganism. Common bacteria affecting GIT are B. cereus, Campylobacter, Salmonella, and the toxin-producing E. coli. Viral gastroenteritis can be caused by norovirus, adenovirus, astrovirus, and rotavirus. Parasitic infections usually affect developing countries and are associated with poor sanitation, e.g., G. lamblia, and Cryptosporidium [62].

The mucosal surfaces represent the primary point of contact for most pathogens attempting to gain entry to a host. Mucosal immunity comprises adaptive and innate immune system mechanisms to counter this. The innate immunity component is exemplified by the mucus secretion system (goblet cells), the barrier of epithelial cells, and the presence of immune cells—mainly phagocytes and NK cells. Following activation of the innate immune system, liaison mechanisms in the form of DCs and macrophages sample luminal antigens and present them to naive T cells in the regional lymph nodes. This causes their cytokine-induced differentiation, activating the adaptive immune system [62].

The mucosal immunity can be protective and detrimental, as illustrated by its effect following H. pylori infection. Specifically, a surge of CD4+ T cells and eosinophils in the gastric lamina propria leads to cytokine release and causes the activation of adaptive immunity [63]. The innate component is primarily dependent on the action of β-defensin. The expression of chemokines, e.g., chemokine (C-X-C motif) ligand 1 (CXCL1) and CXCL2, increases neutrophilic recruitment, leading to improved phagocytosis and active killing of bacteria. Conversely, immune escape will occur through surface antigen switching when the repair mechanism cannot counter the damage. Combined with the accumulation of immune cells and the secretion of their respective cytokines (e.g., IFN-γ, TNF-α), inflammatory responses are promoted, culminating in injury [63].

In the case of parasitic infections, mucosal immunity is mainly carried out by Th2 lymphocytes (with secretion of IL-3, IL-4, IL-5, IL-9, and IL-13), eosinophils and basophils, mast cells, and IgE production. Secretory IgA also plays a vital role by forming complexes with parasitic antigens that have crossed the epithelial barrier [62]. In the case of rotavirus...
infection, mucosal immunity is accomplished by producing specific secretory and fecal IgA, whereas systemic immunity is provided through IgG or IgM [64].

However, these innate immune cells have critical protective roles during infection, such as intracellular pathogen killing. Notably, the disease can cause kidney transplant problems and graft loss due to innate immune cells’ release of inflammatory cytokines [44]. Innate immune memory is not limited to the initial infection (or other inflammatory stimuli); exposure to one pathogen can enhance protection against a subsequent challenge with a different pathogen, even from a different kingdom. “Innate immune tolerance” is another term for innate immunological memory, which is not always expressed as learned immunity [65]. Accordingly, exposure to relatively high doses of bacterial lipopolysaccharide (LPS) in the past by myeloid cells, such as monocytes and macrophages, results in a state of decreased ability to elicit pro-inflammatory cytokines upon subsequent restimulation with LPS (homologous tolerance) or other pro-inflammatory stimuli (heterologous tolerance) [65].

Trained immunity could have evolved as an extension of innate immunological memory to provide extensive cross-protection against re-infections [44]. However, in modern society, trained immunity may contribute to developing age-related chronic inflammatory disorders. Such maladaptive trained immunity may be inappropriately generated by microbial or endogenous stimuli (i.e., DAMPs), resulting in excessive immune responses that cause or aggravate inflammatory or autoimmune disorders. In other words, if the immune system has been epigenetically trained (due to previous infection, vaccination, or injury) to elicit a heightened immune response, this enhanced responsiveness may exacerbate existing inflammatory/autoimmune diseases or be harmful in hosts genetically susceptible to inflammatory/autoimmune diseases. Clinical observations and animal model experiments support these hypotheses [44].

Therapeutically targeting trained immunity is a promising strategy for managing innate immunity and, as a result, adaptive immune responses that are activated and controlled by innate immune cell surface molecules and soluble mediators. Trained immunity can be used to either strengthen the immune response to infections and cancers or to suppress the responses that cause autoimmune disorders and allograft rejection. Trained immunity is regulated at several levels, including ligand–receptor interaction and metabolic and epigenetic regulation [12].

6.3. Influence of Dysregulated Immunity on Gut Disorders

As Levine postulated [66], the gastrointestinal tract has a two-faceted role: a physical barrier and an active immunogenic system. For instance, decreased absorption of pathogens is accomplished through the integrity of the GIT and the complex-forming properties of IgA when encountering a foreign antigen. Conversely, the immunogenic response is activated when antigens bind to the epithelium, triggering a cytokine-mediated inflammatory process [66].

Consequently, dysfunction affecting intestinal tight junctions, immunologic dysregulation, or impaired oral tolerance can potentially promote overwhelming inflammatory responses. Chronification of these processes has been associated with increased susceptibility to autoimmune intestinal disorders [66]. As has already been discussed, ongoing activation of the immune system against intestinal flora seems to be the pathogenic mechanism for IBD. Furthermore, both celiac disease and eosinophilic gastroenteritis have been associated with the breakdown of oral tolerance [66].

Conversely, gut dysbiosis (i.e., altered gut microbiota) may promote systemic inflammation by allowing lipopolysaccharide—an endotoxin found in Gram-negative bacteria—to penetrate the gut epithelial barrier [67]. In the altered microenvironment, increased neutrophilic recruitment leads to collateral damage through cytokine secretion, matrix metalloprotease production, and further immune cell activation. In neonates, gut dysbiosis following formula feeding leads to decreased diversity and expansion of the Enterobacteria-
aceae population. This, in turn, enhances inflammation and promotes the occurrence of necrotizing enterocolitis [68].

6.4. Gut Disorders and Trained Innate Immunity

Most concepts of trained innate immunopathology are related to systemic autoimmune diseases, such as SLE, kidney transplantation, kidney failure, etc. [12]. However, no reliable data on gastrointestinal disorders associated with trained innate immunity were found.

7. Therapeutic Implications for Gut Mucosal Immunity Modulation

According to the Guidelines for Selecting a Therapeutic Approach in IBD (STRIDE-II), continuous disease monitoring from symptomatic relief to endoscopic cure, together with short- and long-term therapeutic responses, are critical to providing IBD patients with a personalized therapeutic algorithm [69].

However, along with therapeutic modulation, we must remember that other factors, such as environmental exposure and diet, and internal factors, such as genetics, sex, race, and aging, could impact mucosal immunity.

IBD treatment has shifted from conventional (aminosalicylates, glucocorticoids, and immunomodulators (thiopurines and methotrexate)) to biologics. Recently, biological drugs containing antibodies targeting specific molecules or proteins involved in the inflammatory process have been developed. Biologic therapies are intended for patients with moderate to severe disease who are unresponsive to conventional treatment [70].

Several biologic therapies are approved for moderate to severe UC and CD, including anti-TNF antibodies and integrin receptor antagonists. The introduction of anti-TNF in the treatment of IBD has shown superior therapeutic effects compared to conventional treatment. Several TNF inhibitors have been developed to control the intestinal inflammation and clinical symptoms of IBD. TNF-\(\alpha\) plays such an important role that agents such as adalimumab, infliximab, certolizumab, and golimumab are used as standard therapy for treating UC and CD [71]. However, anti-TNFs are not effective in all patients with IBD, as 10–30% of patients lack a primary response [72], and another study evaluating long-term outcomes showed that a significant number of patients relapsed after stopping treatment [73].

Developing new targeted drugs aims to improve the clinical effect and reduce systemic side effects. The immunopathogenesis of IBD is complicated by imbalances in different T cell subsets (Treg, NK cells, TH9) and their interactions. As a result, the production of multiple cytokines is impaired. These cytokines include TNF-\(\alpha\) as well as IL-1\(\beta\), IL-6, IL-8, IL-10, IL-12, IL-17, IL-23, and TGF-\(\alpha\) [74].

Several studies have highlighted the role of the IL-23/IL-17 axis in the pathogenesis of IBD. It is known that IL-23 plays an essential role in the regulation of Th17 cells and the stimulation of pro-inflammatory cytokines in patients with IBD [75]. Anti-interleukin inhibitors in development include Ustekinumab, which targets the p40 subunit of IL-12 and IL-23. FDA-approved for the treatment of adult IBD patients with moderate to severe disease. Ustekinumab has shown efficacy in inducing and maintaining clinical remission in patients with active CD and UC [76]. The humanized monoclonal IgG1 antibody Risankizumab, targeting the p19 subunit of IL-23, is in clinical trials. Preliminary results show that the drug is well tolerated and can mediate long-term clinical response and endoscopic remission in patients with active CD [77].

Since Janus kinase (JAK) is vital in modulating the signal transduction pathway of several pro-inflammatory cytokines directly involved in gastrointestinal inflammation, their inhibition also has a substantial therapeutic effect. At this stage in clinical trials, JAK inhibitors have shown good efficacy results in patients with moderate to severe IBD [78]. Tofacitinib is the first-in-class oral JAK inhibitor known to be effective and safe for patients with moderate to severe UC [79]. However, further research on the safety profile in larger clinical trials is needed.
Anti-integrin therapies are novel therapeutics in IBD. They inhibit leukocyte extravasation by blocking the interaction between integrins (cell surface proteins) found in immune cells and adhesion molecules in endothelial cells [80]. The first drug approved by this group for CD is Natalizumab, which acts on α4-integrin. Still, its use is limited due to the risk of progressive multifocal leukoencephalopathy [81].

Vedolizumab affects the intestinal trophic α4β7 integrin, acts selectively on the intestine, and shows few systemic adverse effects with proven efficacy in patients with moderate to severe UC and CD refractory to TNF antagonist therapy. In May 2014, the FDA and EMA approved the drug for treating IBD [82]. Trolizumab, affecting β7-integrin, and PF-00547569, targeting mucosal ICAM-1, are under investigation [80].

Other molecules, new therapeutic approaches, and monitoring of biomarkers to monitor the therapeutic response to different therapies among IBD patients are also under investigation, with an improved clinical effect expected in the future.

8. Challenges and Future Perspectives in Mucosal Immunity and Trained Innate Immunity


Gut immunity has been increasingly used to explain the interplay between gut microbiota and host immune response. As gut dysbiosis is now implicated in the pathogenesis of various autoimmune diseases, including IBD, rheumatoid arthritis, and diabetes type 1, understanding the innate and adaptive immune responses is paramount. However, the dynamic character of these interactions with genetic and environmental factors makes this endeavor quite complex.

The current models for investigating gut immunity involve simulation in germ-free laboratory animals. This clashes with the naturally ‘dirtier’ human microbiome and, therefore, lacks generalization potential [83]. Another limitation of these models is the restricted microbiome composition in both specific pathogen- and germ-free mice, making the results not reproducible in humans. However, the advancement of next-generation sequencing and the fecal microbiota transplant mouse model appears promising in successfully addressing and surpassing those issues [84].

In Table 1, we present models providing various insights into mucosal immunity, each with unique strengths and limitations that influence their suitability for specific research questions.

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<tr>
<th>Model</th>
<th>Characteristics</th>
<th>Advantages</th>
<th>Limitations</th>
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<tbody>
<tr>
<td>Murine Models</td>
<td>Inbred mice strains (e.g., BALB/c, C57BL/6); genetically modified mouse</td>
<td>Controlled genetic background; extensive immunological tools</td>
<td>Differences from human immunity; ethical concerns</td>
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<tr>
<td>Gnotobiotic Mouse</td>
<td>Germ-free or defined microbial communities</td>
<td>Allows study of host-microbiota interactions in a controlled setting</td>
<td>Technically challenging; expensive; limited availability</td>
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<tr>
<td>Humanized Mouse Models</td>
<td>Mice engrafted with human immune cells/tissues</td>
<td>Reflects human immune response; useful for studying human-specific pathogens</td>
<td>High cost; limited lifespan of human cells in mice; complex to establish</td>
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<tr>
<td>Organoid Cultures</td>
<td>3D culture systems derived from human or animal gut stem cells</td>
<td>Mimics human gut tissue architecture and function; high throughput</td>
<td>Lack of complete immune system interaction; variability in protocols</td>
</tr>
<tr>
<td>In vitro Cell Culture</td>
<td>Epithelial cell lines, immune cells (e.g., dendritic cells, macrophages)</td>
<td>Controlled environment; cost-effective; high reproducibility</td>
<td>Simplistic; lacks tissue complexity and systemic immune interactions</td>
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<tr>
<td>Zebrafish Models</td>
<td>Transparent larvae; genetic manipulation possible</td>
<td>Visualize immune responses in real-time; cost-effective</td>
<td>Differences with mammalian immune system; limited antibodies/tools</td>
</tr>
<tr>
<td>Porcine Models</td>
<td>Pigs with similar gastrointestinal physiology to humans</td>
<td>Relevant to human gut physiology; size allows surgical techniques</td>
<td>High maintenance cost; ethical concerns; less developed genetic tools</td>
</tr>
<tr>
<td>Non-Human Primates</td>
<td>Species closely related to humans</td>
<td>Closest physiological and immunological model to humans</td>
<td>Ethical concerns; high cost; limited availability</td>
</tr>
</tbody>
</table>
8.2. Emerging Trends and Future Directions in Research

If elucidated, the intricate relations surrounding the regulation of immune homeostasis can lead to a better understanding of autoimmunity. The concept of trained gut immunity can, therefore, be groundbreaking in two ways: by manipulating innate immune memory using defined therapeutic targets to either enhance innate immune responses for the treatment of infections or malignancies or downregulate excessive inflammation in inflammatory and autoimmune diseases [44].

In this context, as Wu et al. suggested [83], trained innate immunity can serve as the starting point for developing new targets for immunotherapy or expanding the pool of diseases treated with intestinal microbial transplants. Colorectal carcinoma and pancreatic ductal adenocarcinoma are examples of cancers correlated with altered gut microbiome. The subsequent dysbiosis induced PD-1 blockade, bypassing immune checkpoints. In this context, the hypothesis that trained immunity can be manipulated in such a way as to reduce inflammation and reestablish immune homeostasis has tremendous potential.

The role of pre-, pro-, and postbiotics seems to be another emerging topic following the understanding of new aspects of gut immunity. Accordingly, specific strains of *B. fragilis* and *Clostridium* are being investigated for their potential immunosuppressive role in IBD through the induction of Tregs when combined with fecal microbial transplants [84].

9. Conclusions

In conclusion, understanding the intricacies of mucosal immunity and trained innate immunity in the gut is crucial for deciphering the pathogenesis of gastrointestinal diseases and developing novel therapeutic strategies. By elucidating the mechanisms underlying mucosal immunity and the concept of trained innate immunity, we can explore innovative approaches to modulate immune responses and restore intestinal homeostasis in the context of gastrointestinal disorders.

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