REVIEW ARTICLE
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Cellular plasticity of CD4 T cells in the intestine

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Barrier sites such as the gastrointestinal tract are in constant contact with the environment, which contains both bene cial and harmful components. The immune system at the epithelia must make the distinction between these components to balance tolerance, protection, and immunopathology. This is achieved via multifaceted immune recognition, highly organized lymphoid structures, and the interaction of many types of immune cells. The adaptive immune response in the gut is orchestrated by CD4 helper T (Th) cells, which are integral to gut immunity. In recent years, it has become apparent that the functional identity of these Th cells is not as xed as initially thought. Plasticity in differentiated T cell subsets has now been rmly established, in both health and disease. The gut, in particular, utilizes CD4 T cell plasticity to mold CD4 T cell phenotypes to maintain its nely poised balance of tolerance and in ammation and to encourage biodiversity within the enteric microbiome. In this review, we will discuss intestinal helper T cell plasticity and our current understanding of its mechanisms, including our growing knowledge of an evolutionarily ancient symbiosis between microbiota and malleable CD4 T cell effectors.

Keywords: T cells, plasticity, intestines, Th1 cells, Th17 cell

INTRODUCTION

The adult human gastrointestinal tract is the largest surface area of the body that contacts with the environment, covering 200–300 m^2 (1). This intestinal surface is constantly exposed to a diverse range of foreign antigens originating from microorganisms (both commensals and pathogens) and food antigens from the diet (2). Commensal microorganisms play an essential role in extracting nutrients from food that are otherwise inaccessible to the host [such as the metabolism of vitamin K by E. coli (3)], they are required for the development of the host's immune system and for the prevention of colonization of the gastrointestinal tract by pathogens. Mucosal pathogens, including viruses, fungi, parasites, and bacteria, can cause pathology either by local effects after mucosal colonization – such as inducing local in ammation or secreting toxins – or, through systemic infection after breeching mucosa. Microorganism-derived antigens, food-derived antigens, and airborne particles, can be potential immunogens. An inappropriate response to these immunogens at the mucosal surface

The detailed mechanisms underlying T cell plasticity within the GALT remains to be de ned, but several factors that facilitate its occurrence have been proposed and can be divided into extrinsic and intrinsic pathways. In this review, we will summarize the recent literature on CD4 T cell plasticity in the gut, highlight possible underlying mechanisms and discuss its potential benefits for intestinal homeostasis and health.

CD4 T CELL DIFFERENTIATION IN THE GALT

The GALT contains one of the largest lymphoid cell populations found anywhere in the body. GALT is distributed along the intestinal tract and is separated from the luminal content, containing about 100 trillion microorganisms (10) and many dietary products, by a single epithelial layer covered with an intricate network of glycoproteins; the mucous layers. The GALT provides three functions: provides antigenic samples from throughout the GI tract; optimizes the opportunities for naïve lymphocytes to encounter antigen, and nally supports the activated lymphocyte and its initial differentiation. A network of highly organized lymphoid structures comprise the GALT (**Figure 1**) – including mesenteric lymph nodes (mLNs), Peyer's patches (PPs), isolated lymphoid follicles (ILFs), cryptopatches (CPs), and fat-associated lymphoid tissues – as well as the loose connective tissue of the lamina propria (LP). Despite the numerous types of GALT, the organization of all GALT lymphoid tissues shares a basic cellular architecture that facilitates the interaction of APCs with lymphocytes and their subsequent activation and differentiation. However, the GALT tissues differ from each other both in function and in the physical distance to the intestinal lumen; CP and ILFs are located directly underneath the epithelial layer, PPs are further from the lumen and most distant are mLNs and fat-associated lymphoid tissues. The specialized functions of each the GALT tissues have been recently reviewed in Ref. (4), and therefore will not be discussed in detail in this review.

Naïve L-selectin-expressing CD4 T cells migrate from the blood through high endothelial venules (HEVs) to the PPs, CP, ILFs, and mLNs. In these inductive sites of the adaptive immune system, they encounter their cognate antigen presented by APCs on MHC class II. Naïve T cells that did not encounter their cognate antigen leave via efferent lymphatic vessels into the systemic circulation to continue their search for their cognate antigen. In PPs, CP, and ILFs, dendritic cells (DCs) receive antigens transported

some remain in the lymphoid organs to perform their specialized effector functions (11). Upon arriving in the intestine, effector T

(encoding RORγt) and CD161 and showing Treg-typical suppressor activity in vitro. In addition, the majority these FOXP3^{GFP}IL-17^{C} T cells expressed CCR6, a receptor that mediates homing to skin and mucosal tissues (56) and high levels of integrin α4β7 and CD103, markers for gut-homing potential. Analysis of the TCR repertoire suggested that FOXP3^{GFP}IL-17^{C} cells develop from FOXP3^{C} Tregs when exposed to in ammatory signals in the gut (24) and in vitro stimulation of FOXP3^{C} circulating cells can result in IL-17 expression (57

Tregs are more susceptible to colitis and asthma (78). Thus, the gut microbiome is implicated in the protection of autoimmunity and autoinflammation both in the gut and at distant sites. Recent work provides evidence for a symbiosis between CD4$^+$ T cell subsets and the microbiome (28). Lack of Tfr resulted in poor quality IgA production and a limited biodiversity of the microbiome. The presence of Tfr permitted a more diverse microbiome including a larger representation of non-pathogenic Clostridia (28), which establishes a positive feedback loop between CD4$^+$ cells and the microbiome mediated through SCFA, Foxp3 expression, and IgA production. This narrowing of the microbiome and subsequently SCFA deplete environment might explain the association between IgA deficiency and allergy and autoimmunity (66). Taken together, these studies provide evidence that the gut microbiota and CD4$^+$ T cell fate within the gut are inter-dependent, with each affecting the composition of the other.

CD4$^+$CD8a$^+$ CTLs are absent from the intestines of germ-free mice and of mice mono-colonized with segmented filamentous bacteria (SFB), but appear in the intestines following reconstitution with specific non-pathogenic microorganisms (30). Little is known about the mechanisms how antigens originating from microbiota induce a phenotype switch. However, data on Th17 cell differentiation indicate that SFB colonization promotes Th17 cell commitment by the expression of inflammation-associated genes, such as the gene for serum amyloid A (SAA) (79). Some Th17 cells from murine gut have TCR specific for SFB-derived antigen (80). In addition, it has been shown that ATP derived from commensal bacteria activates CD70^{high}CD11c^{low} cells in the LP, leading to enhanced differentiation of Th17 cells (81). Therefore, changes in the concentration of SAA or ATP could either induce a switch toward Th17 phenotype or destabilize Th17 cell commitment toward another phenotype.

DIETARY PRODUCTS

Dietary products, mainly after being processed by microorganisms, can induce phenotype switch. PPARg has been identified to induce a Th17-to-Treg cell switch (23). This nuclear receptor regulates fatty acid storage and glucose metabolism. Therefore, consumption of food containing certain fatty acids may promote a phenotype switch from Th17-to-Treg cells. Several dietary products have been reported to promote or inhibit lineage commitment [reviewed in Ref. (2)]. It is likely that these factors are also candidates to induce a phenotype switch of CD4$^+$ cells. In addition, dietary products could influence a phenotype switch by affecting metabolic and signaling pathways and epigenetic status.

INNATE IMMUNE CELLS: PROVIDERS OF COSTIMULATION AND CYTOKINES

Innate immune cells are abundant in the GALT. They have the potential to influence CD4$^+$ T cell plasticity via their determination of the micro-environment via the secretion of soluble mediators, expression of co-stimulatory molecules, and via their potential to act as APCs. Mucida et al. demonstrated that continuous activation of CD4$^+$ T cells by oral administration of an antigen is necessary for a phenotype switch to CD4$^+$ CTLs (30). It has been reported previously that antigen dose and peptide/TCR affinity can influence Th commitment (82).

Co-stimulatory factors have been shown to play a role in Treg-to-Tfh plasticity. Blocking the interaction of CD40, expressed on APCs such as B cells and DCs, with CD40L, found on T cells, is able to prevent a phenotype switch from Treg cell to Tfh cell (27). The role of cytokines in Th17 cell plasticity has been studied by several investigators but has not yet been fully elucidated. However, it is clear that IL-23-dependent pathway does play an important role. In Helicobacter hepaticus-induced typhlocolitis, mRNA levels for IL-23p19 were elevated after bacterial inoculation and IL-17A$^+$ cells isolated from the colitic intestine expressed both subunits of the IL-23R, indicating that IL-23 acts on Th17 cells to induce a program resulting in IFNg production (18). T cell transfer studies also showed that IL-23 is required for the appearance of IL-17A$^+$IFNg$^+$ double-producing T cells in the intestine. Transferred nai

identity and are likely to play a prominent role in ultimately generating hybrid Th cells and Th identity conversions. Co-expression of IFNg and Th2-associated cytokines in the same T cell seems to be enabled by signals that keep both Tbet and GATA-3 expression in balance, which is in contrast to studies examining chromatin modi cation at Tbx21 and Gata3 (86, 87).

The loss of a single transcription factor can tip the balance in favor of an alternative lineage. In Bcl11b-de cient mice, GATA-3 expression in Th17 cells is unrestrained, resulting in GATA-3-mediated IL-4 production (22). This change in cell phenotype feeds back to the micro-environment, and has further implications for T cell biology. For example, the cytokine mix produced by Th2/Th17 hybrid cells triggers gut-imprinting properties in DCs. IL-4 together with GM-CSF enhance the expression of the enzyme

Rhmin2(g)2(9.464(t)10(o)-2* (0 Td (ldh)3(nme)3(h)2Tj /T(ith)9)-2I3(ip(ith)-7d)-396(c)7(ha(e)- ith)-7d)-3c7P1/T(ith)--2(t)d ()14(o)3(m1

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