university of groningen

Second Research Project

Effector T cells in celiac disease: Comprehensive study on their interaction in chronic inflammation and autoimmunity

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August 2021

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Abstract

Celiac disease (CeD) is a common autoimmune disease, mainly mediated by T cells. The only current therapy is a lifelong gluten-free diet. It involves CD4+ T cell-mediated gluten recognition, and effector T cells, including CD8+ and γδ T cell subsets. Gliadin-derived peptides, derived from gluten, activate lamina propria gliadin-specific CD4+ T cells in a TCR dependent manner, which leads to the release of cytokines. A non-circulating subset of effector T cells called tissue residency memory T cells (TRM) play a crucial role in the

pathogenesis of CeD. These effector T cell subsets produce pro-inflammatory cytokines and cause inflammation and further injury in the intestine.

Celiac disease pathogenesis is also linked to resistance to Treg suppression by the potent cytotoxicity of IL (Interlukin)-15. Furthermore, such resistance may play a role in the development of autoimmune diseases. Effector T lymphocytes from active CeD become resistant to suppression by Treg. This resistance might cause a loss of tolerance to gluten. This role for TRM cells in intestinal autoimmunity raises the possibility for therapeutics directed at resident T cell populations. In this review, we have sought to examine the putative novel treatment to restore anti-inflammatory cytokines and optimal manipulation of Treg suppression and mitigate the inflammatory role of Teff (T effector) cells.

Introduction

CeD is a complex disease that is driven by the effect of environmental triggers, genetics, and local immune regulation. It is an autoimmune disease triggered by gluten intake, characterized by a robust inflammatory response in the small intestine. Even though the primary pathological lesion is in the proximal small intestine, it is a systemic condition. (Jabri and Sollid, 2017).

This disease can also be defined as food sensitivity, in which the body overreacts to gluten in genetically predisposed people. CeD is triggered by the intake of gluten proteins, which are storage proteins. Cereals such as wheat, barley, and rye contain those proteins, in turn composed of gliadin peptides. (Moerkens, Mooiweer, Withoff, & Wijmenga, 2019). The immunopathology of celiac disease results in intestinal disruption by T cells upon sequential responses of immune populations and local inflammation. T cells play a crucial role in such inflammation and autoimmunity, both effector and regulatory T cells working in parallel to obtain an optimised immune response against foreign pathogens and limiting aberrant responses against food antigens. (Khan & Ghazanfar, 2018).

T cells that migrate to the secondary lymphoid organs express a T cell-specific clonal receptor (the TCR) to recognize antigens, and they have two subtypes. The most common is the αβTCR, using α and β chains, which is generally restricted by MHC complexes. The other is the γδTCR, made up of γ and δ chains, which is less well studied, shows more diversity in ligand recognition and restriction, but nevertheless is crucial for primary immune responses. (van Wijk & Cheroutre, 2010; Pennock et al., 2013). Conventional αβTCR T cells are divided into CD4+ and CD8+ subsets. CD4 and CD8 are coreceptors, involved in the restriction of CD4+ T cells to MHC Class II molecules, and CD8+ T cells to MHC Class I molecules, mediateing the antigen-specific activation of effector T cells. Upon encountering an antigen that its receptor can bind, naïve T cells can proliferate and differentiate into one of many forms of effector and memory T lymphocytes.

T cells found in the intestine are generally thought to be protective. However, they might become activated in the absence of a pathogen and this activation may result in autoimmune disease. In this context, celiac disease is considered a clear example, where such uncontrolled activation leads to duodenal inflammation and villous atrophy. (Jabri et al. 2000). In the

following sections, we will decipher the naïve and effector T cells' phenotypes, their functions, and how CD8+ T cell subsets and CD4+ T cell subsets, TRM cells are involved in CeD's immunopathogenesis. Other cell types, including gamma delta T cells and unconventional T cells such as MAIT cells, are also of interest in CeD but are beyond the scope of this review.

Immunopathogenesis of celiac disease

Even though the immunopathogenesis of CeD is well defined, there are still gaps in our knowledge. Although carrying HLA-DQ2 and HLA-DQ8 alleles in terms of genetics is an essential factor for the formation of celiac disease, it is not sufficient to cause the disease. (Withoff, Li, Jonkers, & Wijmenga, 2016). Broadly, the model of coeliac disease immunology is represented in Figure1.

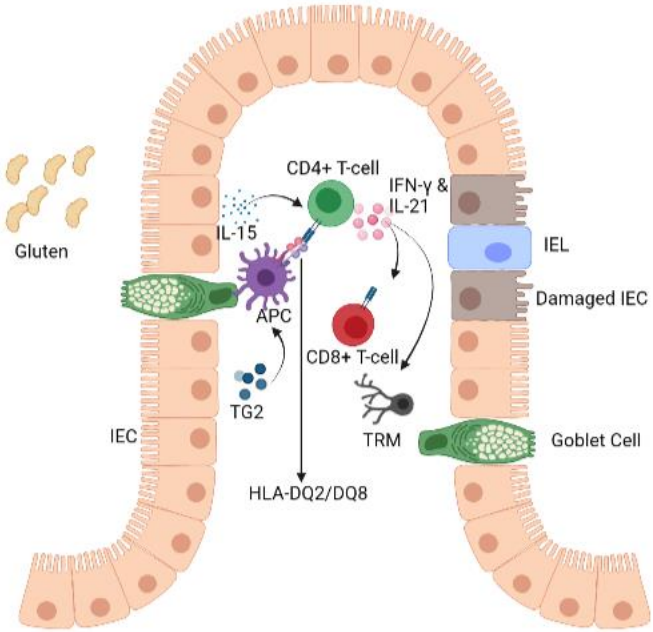

Figure 1 Immunopathology of CeD

Firstly, semi-digested gliadin peptides from gluten may bypass the intestinal epithelial (IEC) and enter the lamina propria (LP).

Here, the peptides are deamidated by Tissue Transglutaminase 2 (TG2), and they are taken up by antigen-presenting cells (APCs). APCs present the deamidated gluten peptides to CD4+ T-helper (Th) cells.

Upon recognizing gliadin antigen, CD4+ Th cells secrete specific cytokines, such as IL-21 and IFNγ. Those cytokines facilitate the activation of intraepithelial lymphocytes (IEL).

CeD is characterized by a cytokine response of IELs to gliadin antigen and an increase in pro-inflammatory IFN-γ.

IL-10 is downregulated in the pathology of active celiac disease, without an increase in tumor necrosis factor-α (TNF-α) or transforming growth factor-β1 (TGF-β1). (Forsberg et al., 2007). Subsequently, IELs are recruited to the intestinal epithelium, and after their massive expansion, they attack the epithelial cells, leading to villous atrophy. (Barone, Troncone, & Auricchio, 2014) (Mayassi & Jabri, 2018).

Upregulation of IL-15 in the epithelium and the intestinal lamina propria (LP) also correlates with suppression of Treg cells and leads to mucosal damage. (Abadie & Jabri, 2014). CeD is mainly characterized by villous atrophy, and it leads to malnutrition of patients due to the low absorption of nutrients by the impaired surface in the small intestine.

As can be seen from the above description, the key T cell subsets are all local to the duodenum. In the next sections, the development of local T cell immunity focusing on resident memory cells is described.

Understanding the Role of T cells

Understanding the role of the effector T cells in celiac disease is subcategorized as following,

- Introduction to subgroups of TEM, TCM and TRM, with comprehensive discussion on TRM, concerning its direct correlation with CeD.
- CD8+ T-cell, with its deficiency linked to chronic autoimmune diseases and its significant presence in epithelial barrier sites.
- Role of the intestinal epithelium (IEL) found in the mucosal lining, defined as the antigen-experienced T cell subset.
- CD4+ T-cell, being the source of many key cytokines and importance in understanding of its adaptiveness to immune response.
- Regulatory T cells (Tregs) and their function in suppressing the immune response, with discussion of putative treatment techniques concerning Tregs.
- Discussion of memory T cells with their phenotypic marker expression to evaluate population, function and localization.

Naïve T cell conversion to Effector and memory T cells

T cells that have never been exposed to an antigen are known as naive T cells (TN), and when they are exposed to an antigen, they become antigen-experienced T cells. This includes effector cells which can elicit rapid and efficient protection against intruders, as well as longer term memory subsets. (Wu et al., 2018). Memory T cells have been traditionally grouped into two major subsets: effector memory cells (TEM) that are enriched in blood and central memory T cells (TCM) mostly found in the lymph nodes (Thome et al., 2014).

The other subgroup, tissue-resident memory T (TRM) cells provide long-lived protective immunity as a subset of memory T cells. As its name suggests, they are non-circulating memory T cells and reside in tissue. (FitzPatrick et al., 2021). They are localized in nonlymphoid tissues (NLT) and secondary lymphoid organs, and they became permanent in NLT. (Masopust et al., 2010). TRM cells are the significant guardian and provide local immune surveillance by

generating in situ, adaptive protection in infections. However, they can also drive immunopathology of the diseases and autoimmunity. (Sasson et al., 2020; Steinbach et al., 2018). TRM cells could emerge from its bystander activation as allergen-reactive or self-reactive T cells. Bystander activation is defined as an antigen-independent activation that could be observed in inflammatory environments, including autoimmunity. (Whiteside et al., 2018). Hence, it contributes to chronic inflammation. (Bartolomé-Casado et al., 2019). To identify the clinical benefits, including the novel treatments for chronic inflammation, the phenotype, location, and maintenance of TRM cells should be deciphered.

It is therefore summarized that the TRM cells are usually confined to mucosal sites, such as the intestine and not in blood, hence it is a great concern in the understanding of CeD. (Sheridan & Lefrançois, 2011).

TRM function

CD8+ TRM cells can generate an antiviral and antimicrobial environment by secreting IFNγ and TNFα. (de Leur et al., 2019). CD69+ T cells produce elevated amounts of the pro-inflammatory cytokines IFN-γ, IL-13, IL-17A, and TNF-α, linked with chronic inflammation in the intestine. (Samat et al., 2021). Moreover, $CD8^{+}CD49a^{-}$ TRM cells, found in mucosal tissues, can produce cytokines such as IL-17 and IL-22 production. (Schenkel et al., 2014; Schreurs et al., 2021). However, inflammatory signals found in the intestine could change the path of the T cells differentiation. IL15 overexpression also activates CD8 + T cells in the intestine. Moreover, IL15 is critical for the TRM cells' long term maintenance. (Samat et al., 2021; Jabri and Sollid, 2017). Protection of the host tissues entails the interaction between TRM cells and innate and adaptive leukocytes. TRM cells also recruit other immune cells. They secrete IFN-g mediates endothelial upregulation of vascular cell adhesion molecule 1 (VCAM-1). It recruits circulating memory $CD8^{+}$ T cells and B cells to sites of the second activation of TRM. (Schenkel et al., 2014b). Functionally, the majority of tissue-resident T cells were quiescent (Sathaliyawala et al., 2013). Adult intestinal TRM cells correspond to a higher differentiated phenotype. (Schreurs et al., 2021). Tissue memory T cells have a heightened activation state that can be satisfied by responses to low antigen doses. (Sathaliyawala et al., 2013). Intraepithelial cytotoxic lymphocytes, which require recognition of low affinity antigen to kill the epithelial cells or lower the T cell threshold for activation, could efficiently function and secretes IFN g . IL-15 can also reduce the TCR activation threshold.

Table 1 The phenotype and construction of intestinal TRMs, to clarify TRM signatures, phenotypic and transcription factor, proliferation survival regulation, and functions.

TRM Marker			
Surface markers	**Function**	**Correlated Cytokines and chemokines that are involved in constructing TRM population**	**Roles of chemokines and cytokines**
CD69	Antagonisation of S1P1-mediated tissue egress	IL-15, IL-2, TGF β,	receptiveness to TGF-β for proper T_{RM} formation and function, expansion (Topham & Reilly, 2018; Raeber et al., 2018)
CD103	Localization via binding to E-cadherin (Iijima & Iwasaki, 2015)		
CD44	Binding to hyaluronic acid localization and/or maintenance, represents previous infection (Masopust et al., 2010).	CCR7Low	T cell trafficking and compartmentalization within secondary lymphoid organs (Bromley et al., 2020)
CD49 a	Binding to collagen and laminin and specialization effector function (Bromley et al., 2020)		
CD127	Homeostatic proliferation (Martin & Badovinac, 2018)	CXCR3	integrin activation chemotactic migration (Iijima & Iwasaki, 2015)
CD62 L low	Adhesion molecule and low level represents the prevention of tissue exist(Masopust & Soerens, 2019)	Granzyme B	Cytotoxicity (Raeber et al., 2018)
CD244	Signaling lymphocyte activation molecule (Bergsbaken et al., 2017)		

Transcriptional regulators	
downregulation of HOBIT	Tissue retention (Mami-Chouaib et al., 2018)
downregulation of T-bet	increased TGF-β responsiveness
Upregulation of Notch	maintenance of CD8 T_{RM}
Ki67	Proliferation marker
down-regulation KLF 2 ($KLF2^{lo}$)	to prevent S1P1 expression & maintenance in tissue (Kim & Harty, 2014)

TRM cells could cause a chronic inflammation

Recent studies demonstrate that the presence of TRM cells corresponds to chronic inflammation and autoimmune diseases. (Steinbach et al., 2018; Mayassi et al., 2019; Zundler et al., 2019). TRM cells can be activated in the barrier tissues and cause an accumulation in response to antigens, which results in immune inflammation, which could later convert into persistent inflammation by activation in other than barrier tissues. (Park & Kupper, 2015). This correlation could be attributed to TRM cell persistence due to longevity rather than *in situ* proliferation (Thome et al., 2014). There are several diseases associated with the presence of cytokine secreting TRM. Psoriasis is an IL-17-mediated autoimmune disease. It is characterized by recurrent skin lesions in the exact locations, which indicate the demonstration of tissue-resident T cells. $CD8^+$ TRM cells have been proved to secrete IL-17a or IFN-gamma, and $CD4^+$ TRM mediates IL-22 in psoriatic skin. Multiple sclerosis (MS) is also an autoimmune disease caused by TRM cells in brain tissue. (Wu et al., 2018)

TRM cells could also modulate the migration and differentiation of other immune cells into the barrier tissues. Therefore it has a crucial role in balancing the immune response. Consistently, a high proportion of TRM cells in the intestine could cause chronic inflammation, such as IBD. (Bottois et al., 2020). The production of IFNγ by TRM is also associated with alopecia areata. Vitiligo is also associated with TRM cells and its secretion of perforin and granzyme B. (Sasson et al., 2020). TRM cell deficiency has also been linked to lupus, type 1 diabetes, and rheumatoid arthritis.

The emerging role of TRM cells in CeD

Human intraepithelial cytotoxic lymphocytes (IE-CTLs), also known as oligoclonal antigen-driven tissue resident T cells, are playing an essential role in celiac disease. (Hayday, 2000). Human IE-CTLs are Ag-driven, highly oligoclonal effector T cells that express CD69 and the integrin CD103. (Jabri & Abadie, 2015) Those IE-CTL react to gluten peptides, produce IFN g, and it is claimed to kill epithelial cells based on recognition of stress signals. (Bergsbaken et al., 2017). It is likely to an indicative of TCR-independent mechanisms are critical CD8+ T cell responses in coeliac disease

In contrast, it is delineated that IE-CTLs mediate tissue inflammation after CD4+ T cell activation in the lamina propria, and IE-CTLs can kill the epithelial cell without encountering gluten. This correlation could be attributed to the patients on a gluten-free diet could display the activity of IE-CTLs in response to the stress signals. The essential cells that play a pivotal role in the immunopathogenesis of CD are also tissue resident T cells. Additionally, TRM cells also suppress the influx of circulating T cells. (Hayday et al., 2001; Kutlu et al., 1993). Hence, they are also involved in the immunopathogenesis of CD indirectly. It seems that IE-CTLs destroy distressed epithelial cells by recruiting the stress signals independently from their TCR specificity, concordant with the notion of villous atrophy caused by the tissue resident cells function, which is not dependent on gluten specificity. Elevated levels of several cytokines and chemokines are correlated with celiac disease. In support of this concept, TRM cells could cause the production of higher levels of chemokines and cytokines by recruiting them.

Putative therapies for TRM in CeD

Intestinal dendritic cells in the presence of IL-15 abrogates the tolerance to dietary Ags and promote T_H1 TRM immune responses against dietary Ags. However, TRM cells elicit local immunization. It seems, therefore, systemic immunization didn't egress. (Depaolo et al., 2011). In other words, systemic immunization that drives protective immunity against mucosal pathogens is less effective than local immunization. (Mueller & Mackay, 2016)
Altogether, it indicates that even TRM cells could target a new therapeutic approach in celiac disease, we could only prevent its activity locally. Moreover, IL-15 lowers the TCR activation threshold and endow CTLs with the ability to kill targets. To block IL-15 cytokine production reduces the resident T cell activity involving the IFN g secretion. Hence, solve the villous atrophy. Blockade of IL15 using monoclonal antibodies has been reported to be of value in treating patients with autoimmune disorders.

Putative therapies that aim to suppress T cell function do not alter T cell localization. Effector progeny that is trafficked back to intestinal mucosa reactivate TRM cells by reacquiring its phenotype. (Casey et al., 2012). Therefore, the disease might relapse. (Park & Kupper, 2015). To establish the desired subset via inhibition, we should recapitulate TRM cells' role in protective immunity. However, tissue resident T cells' function should maintain their balance since they also have a protective role in general immunity, besides their pathogenic role in Celiac disease. Otherwise, blocking TRM cells could result in flushing pathogenic TRM out of tissues and prevents their protective physiological function. (Wu et al., 2018)

TRM Precursors, Development, Maintenance and Phenotypes

CD8+ cells that express low KLRG1, and high CD127, are accepted as the precursor of TRM (Debes et al., 2005). As they are exposed to antigens and cytokines, they differentiate into TRMs (Samat et al., 2021). TGF- β plays an important role here by downregulating T-bet and Eomes expression and upregulating CD103 (Samat et al., 2021). In the development of TRMs, chemokine receptor CXCR3 is also important as it recruits the precursors of TRMs to pro-inflammatory microenvironments, which are favorable to gaining TRM phenotype.

TRMs exist in tissues, with an even higher population in the barrier tissues such as skin and gut. (Kumar et al., 2017). Therefore, tissue-resident memory T (TRM) cells are characterized by their surface markers CD69 and CD103. While CD69 upregulation differentiates all (TRM) cells from circulating T cells, not all TRM cells express CD103. Thus they are subdivided into CD103+ and CD103− TRM cells. (Bergsbaken and Bevan, 2015; Sathaliyawala et al., 2013). ITGAE encodes CD103 differentially regulated in $CD103^{+}$ and $CD103^{-}$ TRM subsets. (Bergsbaken et al., 2017)

However, local tissue infection also drives CD69 expression. CD69 is upregulated on activated effector cells that encounter their cognate antigen. Its expression prevents T cells from leaving the lymph node, but the constitutive expression is specific for resident cells. (Topham & Reilly, 2018). Cytokines, together with the tissue environment, could modulate the activation of the CD69 marker. Additionally, CD69 expression could regulate the secretion of IFN-γ, IL-17.(Cibrián & Sánchez-Madrid, 2017).

Consistently, TRM comprises subpopulations that display differences in different tissue compartments. (Samat et al., 2021). Mucosal T cells, both intraepithelial and lamina propria lymphocytes (LPLs), showed a constitutive expression of CD69 and granzyme B. Thus they are considered as activated effectors until the discovery of TRM cells. (Montufar-Solis et al., 2007; Bergsbaken et al., 2017; Bartolome´ -Casado et al., 2019). TRM cells resemble TEM cells in terms of their phenotype and differentiate from effector T cells by the CD62L expression. TRM expresses CD44 and low levels of CD62L. TRM is located at the initial infection site, providing a very local immune response and protection against reinfection. (Masopust et al., 2010). TRM cells could also compete with innate or innate-like lymphocytes responsible for immune surveillance at the site of the previous infection. (Konjar et al., 2017). The mechanism behind the T cells locking into tissue is the amalgamation of the consecutive stimulation processes. E-selectin, which is expressed by endothelial cells, binds to CD8+ Teff cells, CD8+ Teff cells move to the location of the inflammation. In cohesion, CD4 Teff enters the tissue upon infection, and they secrete IFN-gamma. In conjunction with this, dendritic cells induce secretion of CXCL9 (C-X-C Motif Chemokine Ligand 9). Type I interferons induce secretion of CXCL10, and they recruit CD8 Teff in combination with the process. TGF-ß signal triggers TRM for CD103 expression. CD103 binds to E-Cadherin, consecutively TRM precursors induce CD69 expression, it has proven that CD69 activation suppresses S1P1, which then TRM binds to S1P1 (sphingosine-1-phosphate). (Iijima & Iwasaki, 2015). Hence, functional tissue residency is mediated by loss of S1P1 and downregulation of the transcription factor KLF2. KLF2 is the transcription factor required for the expression of S1P1 and CCR7. CD69 antagonizes S1P1

mediated tissue egress, preventing very recently activated T cells from leaving the secondary lymphoid organ before they become primed T cells. (Casey et al., 2012;Steinbach et al., 2018) As a result of the T cells' locking into the tissue, CD103 (integrins E and 7) expression, sphingosine-1-phosphate receptor 1 (S1P1) antagonist CD69, collagen-binding CD49a), and hyaluronic acid (HUA) binding CD44 markers are all used to identify TRM. CD49a interacts with Collagen, and CD103 adheres to E-cadherin, facilitating the position of the lymphocytes near the epithelial surface. However, TRM cells display heterogeneity in their phenotypic markers. (Topham & Reilly, 2018).
β2-integrin might be an adjunct surface marker for CD103 TRM cells to differentiate them from effector phenotype but also subdivided population depends on CD103 expression. (FitzPatrick et al., 2021). In this review, we will focus on the TRM cells in the small intestine.

Gut homing marker of TRM cells

$CD8\alpha\beta^{+}TCR\alpha\beta^{+}$ memory T cells residing in the intraepithelial lymphocyte (IEL) and lamina propria (LP) compartments of the intestines.
On T cells, the α4 integrin subunit associates with β7 subunits to form $\alpha_4\ \beta_7$ integrin that facilitates gut-homing. Consistently, memory $CD8^{+}$ T cells express $\alpha_4\beta_7$ in the small intestine lamina propria, but $\alpha_4\ \beta_7$ integrin is not necessary for long-term maintenance. (Casey et al., 2012). Within the small intestine, most memory $CD8^{+}$ T cells and colon are CD103+. (Jabri & Solid 2017). Most of the $CD69^{+}$ tissue-resident memory T cells expressing IL-7 receptor (CD127) in mucosal sites. (Steinbach et al., 2018). $CD69^{+}$ populations showed higher expression of Ki67 and BCl2, facilitating proliferation and survival factors of lymphocytes. (Masopust et al., 2010). TRM cells' capacity of responding to IL-12R or being recruited to CXCR3 local inflammatory loci controls the formation homeostasis of the gut. (Y. Liu et al., 2018)

TRM cells in both the intraepithelial and lamina propria compartments of the jejunum displayed high levels of CCR9, which is a chemokine receptor presented on subsets of memory $CD4^{+}$ and $CD8^{+}$ lymphocytes CCR9 expresses the intestinal homing receptor. The ligand for CCR9, CCL25, is essential for migrating lymphocytes into the small intestine. (Cibrián & Sánchez-Madrid, 2017). TGF- β R induces CD103 upregulation in the small intestine. (Casey et al., 2012;Bergsbaken et al., 2017).However, IFN ß and IL-12, produced in intestinal pro-inflammatory microenvironments, suppress TGF- β induced CD103 expression. (Bergsbaken et al., 2017)
TGF- β induces expression of $\alpha_4\beta_7$ on TRM cell precursors. However, innocuous antigens could interfere with $\alpha_4\beta_7$ upregulation. (Bergsbaken et al., 2017). Intestinal $CD103^{+}$ T cells display higher expression of the surface marker CD161 C -type, lectin-like receptor. (FitzPatrick et al., 2021). TRMs in the LP compartment do not express CD103, which indicates that TGF-β elicits TRM cells' essential functions, such as migration towards the gut via CD103-independent mechanism. (Y. Liu et al., 2018). Additionally, CD103 itself does not play a role in TRM retention. (Sheridan et al., 2014)
Integrin $\alpha_1\beta_1$ is another marker highly expressed on TRM in the small intestine. (Gebhardt et al., 2009; Mackay et al., 2013; Ray et al., 2004; Wakim et al., 2012). L-selectin (CD62) antibody marker also identifies the intestinal tissue resident phenotype of conventional lymphocytes. (McDonald, Jabri, & Bendelac, 2018). There are tissue resident CD4+ T cells in barrier tissues.

Human Th17 TRM cells have been characterized in the patients' lamina propria with a signature of inflammation in their intestine. Inflammation cause to release of IL-17 and IFNγ by Th17 TRM. (Schreiner & King, 2018)

CD8 T cells' Phenotypes, Function & Development

CD8+ T cells recognize peptides bound to MHC Class I (represented in Figure 2).- CD8+ αβ+ are a conventional type of T cell; they are also known as cytotoxic T cells and typically have a high expression of granzyme and FasL, which induces cytotoxicity. (Sun et al., 2015). CD8+ T cells typically recognise antigens derived from cytosolic proteins (e.g. endogenous proteins or viruses). The proteasome degrades these cytosolic proteins and CD8 +T cells recognize cognate antigens displayed on MHCI. (Konjar et al., 2017; Pennock et al., 2013) (see Figure 2.)

Besides the protective role of CD8+ T cells in immunity, they are a significant effector in cell and tissue damage. They can kill the pathogen-infected host by cytokine secretion. Those cells gain their effector capacity upon encountering the cognate antigen, and they are considered the subset of type 1 immunity due to their capability of interferon (IFN)γ production. (Konjar et al., 2017). As described above antigen experienced naïve CD8+ T cells differentiate into TM of which there are three major subsets: TEM, TCM, and TRM cells. (Casey et al., 2012; Sathaliyawala et al., 2013). (See Figure 2).

CD8+ T cells' regulation in several diseases illustrate their dual role in immunity. They protect the host from foreign intruders' infection by maintaining tissue homeostasis and integrity; while, they can also sustain inflammation once a perpetual cycle of inflammation has been initiated. (Samat et al., 2021). CD8+ T-cell deficiency has been linked to chronic autoimmune diseases, including multiple sclerosis, rheumatoid arthritis, systemic lupus erythematosus, Sjögren's syndrome, systemic sclerosis, ulcerative colitis, Crohn's disease, psoriasis, vitiligo, alopecia areata, type 1 diabetes mellitus, Hashimoto's thyroiditis. (Pender, 2012).

Antigen-specific CD8 + T cells, if left unchecked, could contribute to chronic inflammation and immunopathology. (Khan & Ghazanfar, 2018). T cell responses decline when the relevant antigen is eliminated to avoid constant inflammation and re-establish homeostasis. (Steinbach et al., 2018). When T cells are deprived of the stimuli secreted during inflammatory reactions to antigen, 95% of effector T cells undergo apoptosis. (Konjar et al., 2017). However, some memory cells are long-lived and maintain protection against reinfection. Memory CD8+ T cells are a heterogeneous population, varying in phenotype, function, and localization. (Konjar et al., 2017; Pennock et al., 2013)

CD8+ TRM cells exist in all tissues, but especially are of significance at epithelial barrier sites such as the skin, lungs, and gastrointestinal tract, those parts that are exposed to antigens frequently.

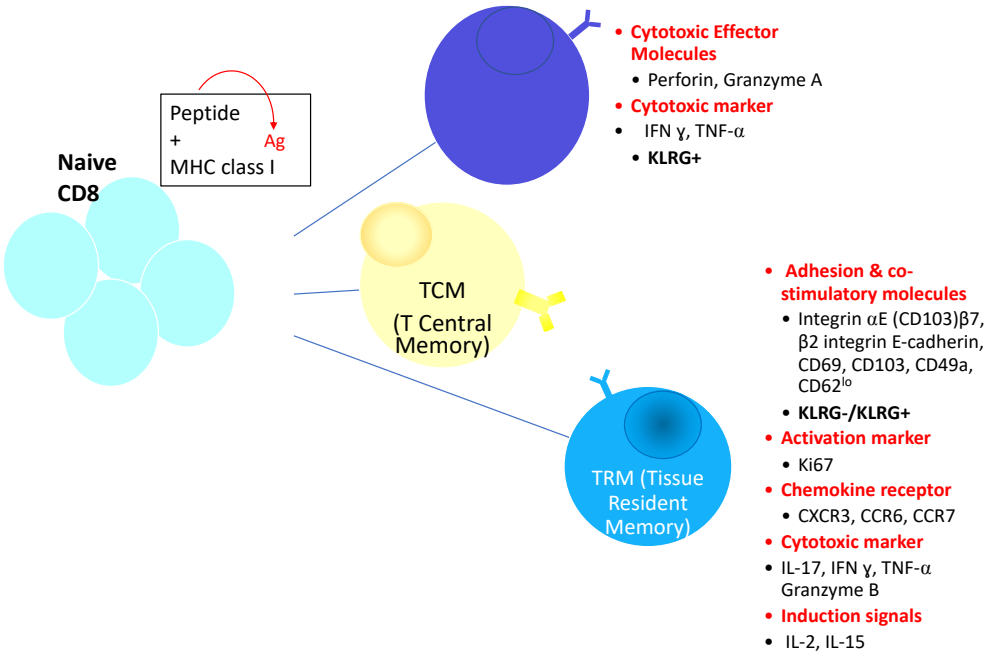

Figure 2 CD8 T cell's differentiation and CD8 TRM and TEM markers CD8+ T cells are differentiated into TCM, TEM, or TRM cells upon encountering an antigen displayed on MHCI. CD8+ TRM cells could produce IL-17, IFN-γ, and Granzyme B as a cytotoxic marker. Ki67 is a proliferation-associated marker.IL-2 and IL-15 promote TRM generation and maintenance, respectively. CD8+ TRM surface molecules are listed as CD103, CD69, CD49a, $CD62L^{lo}$, β2 integrin.

CD8+ T cells in the intestine – IELs

The intestinal epithelium (IEL) is in contact with trillions of microorganisms. Hence, there is a large number of immune cells. (Konjar et al., 2017; Sheridan & Lefrançois, 2010). A large proportion of T cells in the intestine are IELs (intraepithelial lymphocyte); IELs are defined as the antigen-experienced T cell subset found in the mucosal lining.

IELs are an understudied subset but contain a range of different T cells – which also differ between species. These distinct subsets include $TCR\alpha\beta^{+}CD8\alpha\beta^{+}$, $TCR\alpha\beta^{+}CD8\alpha\alpha^{+}$, $TCR\alpha\beta^{+}CD4^{+}$, $TCR\gamma\delta^{+}$. (Sánchez-Castañon et al., 2016). $TCR\alpha\beta^{+}CD8\alpha\beta^{+}$ IEL reside in between the basement membrane and enterocytes, and they comprise conventional and unconventional T cell subtypes. (Sheridan & Lefrançois, 2010; Winter & Krueger, 2019).

$TCR\alpha\beta^{+}$CD8+ αβ+ are a conventional subtype of IEL (Abadie & Jabri, 2014; McDonald et al., 2018), and they are tissue resident memory T cells.(Jabri & Abadie, 2015). Upon encountering the cognate antigen, they could modulate the immune response and repair the epithelial barrier. (Dahan, Roth-Walter, Arnaboldi, Agarwal, & Mayer, 2007; Mowat, 1990). CD8αβ+ T cell priming with its cognate antigen occurs predominantly in the lymphoid organs. In the inflammatory environment, antigen-specific CD8αβ+ T cells undergo expansion, emigrate from priming, and infiltrate effector tissues. During this process, CD8αβ+ T cells acquire homing receptors, which facilitate their migration to the intestine. (Sheridan & Lefrançois, 2010).

Additionally, IELs include a set of T cells marked by expression of CD8αα. This is one feature which differentiates IELs from peripheral T cells. (Ma et al., 2021). The majority of IELs in mice

express the CD8αα homodimer, a marker of T-cell activation. (Sun et al., 2015) CD8αα+TCRαβ + IELs complete their development in the gut. MHC class I molecule's α3 conserved domain and the α chain of CD8 binds to each other. However, the β chain is crucial to confer activity as a coreceptor of the TCR. (Ma et al., 2021). CD8αα appears to function as a TCR corepressor. (Sun et al., 2015). T-bet, which Tregs also express, plays a central role in the formation of the CD8αα+TCRαβ + IEL subset.

CD8αα+TCRαβ + IELs are considered as the intestinal epithelial "Tregs." They undergo thymic agonist selection, which enables them to develop mucosal tolerance. Additionally, CD8αα is incapable of promoting the positive selection of MHC class I-restricted thymocytes. (Cheroutre et al., 2011; Sun et al., 2015). Additionally, interleukin 10 (IL-10), an anti-inflammatory cytokine, is crucial for maintaining normal tissue homeostasis and preventing host tissue destruction. CD8+ T cells produce IL-10 in an attempt to counteract the inflammatory function themselves. (Forsberg et al., 2007)

IELs/CD8+ T cells regulate celiac disease activity with the help of cytokines

Memory CD8+ T cells within the gut epithelium encompass IELs. On the other side, within the IEL compartment, most T cells are CD8+, as either conventional CD8αβ heterodimer or as a CD8αα homodimer. (Shale et al., 2013)CD8+ T cells within the gut epithelium are functionally distinct from CD8+ T cells in lymphoid tissue and exhibit immediate cytotoxicity upon reinfection (Masopust et al., 2010). We will focus on IELs due to the linkage between the increased number of IELs and active CD. (Konjar et al., 2017; Sollid, 2004). IELs are critical players in the formation of villous atrophy. IFN-γ production from the gluten-specific CD8+ T cells is the leading cause of mucosal intestinal destruction. (Konjar et al., 2017; Mazzarella et al., 2008).

In CeD, natural killer (NK) cell receptors and IL-15 drive tissue distress, in which natural killer cells are activated by the upregulation of cytokines and chemokines, such as IL21, CCR9, and RGS1. Those cytokines are also involved in the migration of IELs to the site of inflammation. Following this, IELs destroy distressed intestinal epithelial cells, independently from their TCR specificity. This observation indicates that NKRs and IL-15 can lower the TCR activation threshold, and it can promote CD8+ T cell activation in the absence of co-stimulation. Moreover, IL-15 enhances TGFß activation, which induces CD8+ TRM generation by the downregulation of the T-bet gene. Expression of the proliferation-associated marker Ki67 by the IELs is also shown in active CeD patients.(Halstensen & Brandtzaeg, 1993; Samat et al., 2021). Consequently, massive expansion of CD8αβ TCRαβ IELs via the NK cell receptors is associated with CeD. (McDonald et al., 2018). Furthermore, IL-15 and IL-21 expression synergistically promote CD8+ T cell activation, expansion, memory cell formation and also their cytotoxic capabilities. (Abadie & Jabri, 2014, Ma et al., 2021). In addition, IL-15 and IL-21 secretion by CD8αβ +TCRαβ + IELs drives the resistance of CD4+ T cells to Tregs suppressive effects. Other key cytokines that drive T-cell activation in CeD include IFN-γ and IL-2.

Overall, the dysregulation of certain inflammatory cytokines promotes the overactivation of T cells in CeD patients and thus, they are potential therapeutical targets. Therapies that already target these cytokines in different diseases could also be used for CeD patients to a great effect.

Putative treatments addressing CD8+ T cell activity for CeD

CD8αβ +TCRαβ + IELs are the leading cause of cytotoxicity; they could cause self-injury when the intestinal immune tolerance is impaired. (Ma et al., 2021) Therefore, inhibition of IFN-γ-producing effector memory $CD8^+$ T cells and particularly CD8αβ TCRαβ IELs in mucosal sites could be a putative treatment for CeD. (Sheridan & Lefrançois, 2011).

Since CD8αβ +TCRαβ + IELs are essential for immune homeostasis in the gut, targeting them could however result in detrimental side effects. According to Samat and his colleagues, focusing on blocking the migration of these immune cells to the gut or the inflammation site could lessen the stimulating side effects. They suggest that Vedolizumab, which blocks the gut-homing α4β7 integrin, could be a therapy for Crohn's Disease because it effectively blocks IEL expansion. (Samat et al., 2021). Since such an approach would alleviate the risks of targeting CD8αβ +TCRαβ + IELs and still target the one of the major players of CeD, it is very promising.

IL-2 promotes memory CD8αβ TCRαβ IEL expansion. Hence, IL-2 blockade might be another intriguing modulation to prevent CeD. IL-2 blockade might also be a treatment for autoimmune disease in a broader perspective.. (Apert et al., 2018).

IL-10 production, with its potent anti-inflammatory properties, could compensate for the pro-inflammatory cytokines. Hence, it is inhibitory of inflammation in an autocrine fashion. As a comprehensive case that asses the correlation between IL-10 and autoimmune disease is: the CD8αα+TCRαβ + IELs prevent colitis in the intestine IL-10-dependent manner. (Forsberg et al., 2007). In principle, CeD could also be reverted by inducing CD8αα+TCRαβ + IELs, which play the regulatory role in the intestine, in contrast, to the normal effector function of T cells. (Ma et al., 2021).

Finally, the inhibitory impact of CD8αβ TCRαβ IELs on the Treg cell function raises another possibility for therapeutics directed at the CD8αβ TCRαβ IEL population. IL-15 signaling, which is known to trigger induced $CD8\alpha\beta^+$ IELs, could prevent exacerbation of CeD. (Abadie & Jabri, 2014; Cheroutre et al., 2011).

CD4+ T cells

This section will discuss what induces effector CD4+ T cells to exit secondary lymphoid tissues and move to infection sites, which causes them to leave secondary lymphoid tissues, how expression receptor changes during the adaptive immune response.

Depending on the signals CD4+ T cells receive during priming, they may form various effector and memory cell types. CD4+ effector T cell activity may include cytotoxicity, but their central role is to release cytokines that guide the cells toward pathogen-specific sites or promote B cell maturation. (O'Shea & Paul, 2010).

Additionally, conventional CD4+ T cells may acquire inhibitory potential, described as Treg cells. (Pabst & Bernhardt, 2013). CD4+ T cells or helper T cells have high plasticity. (Korn et al., 2009; O'Shea & Paul, 2010). Differentiated CD4+ effector T cells are accepted as the source of many key cytokines, which play an essential role in adaptive immunity.

CD4+ T cells' activation and function

CD4+ T cells are categorized under the type of cell-mediated immunity, in which the detection of the intruder by the T cells inducts pro-inflammatory cytokines. (Shale et al., 2013). CD4+ T cells express surface molecules and secrete cytokines that activate other effector cells that eliminate pathogens (Seder & Ahmed, 2003). Peptides obtained from protein antigens are presented by dendritic cells in peripheral lymphoid organs, and they are recognized by CD4+ T cells presented on MHC Class II molecules. (Zhu et al., 2010). CD4+ T cells generate IL-2, which helps CD8+ T cells to proliferate. Hence, CD4+ T cells are known for their orchestrating role in the immune response. (Boyman & Sprent, 2012; Williams et al., 2006).

Naïve CD4+ T cells differentiate into effector T-helper cells via activation signals. Activation and differentiation require three distinct signals. In conventional T cell activation, a CD4 coreceptor binds to the foreign-antigen: self MHC II complex, and signal is delivered to the T cell that antigen encountered with previously. (Whiteside et al., 2018) Secondly, the costimulatory signals such as B7 and CD28 of the same antigen-presenting cell is required to activate naive T cells. CD4+ T cells improve the effector functions of other pathogen-killing cells.

Depending on the third signal delivered by the antigen-presenting cell, various differentiation pathways generate distinct Teff subsets (Whiteside et al., 2018). The main CD4+ T eff subsets are T Follicular helper cells, T regulatory cells. (O'Shea & Paul, 2010) together with Th1, Th2 cells, Th9, and Th17 cells, depending on the cytokines they produce. (Konjar et al., 2017; Korn et al., 2009; Lutter et al., 2018; Zhu et al., 2010). In addition to the cytokines, the differentiation of inflammatory CD4+ T cells requires specific transcription factors. (Kanno et al., 2012) (Jabri and Sollid, 2017).

CD4+ T cells in the intestine – Tregs

Mucosal tissues are constantly exposed to exogenous materials such as dietary components and microorganisms. Therefore, they have a high amount of CD4+ T cells. (Sathaliyawala et al., 2013; Shale et al., 2013) As discussed in the TRM section, tissue-resident T cells are mainly quiescent or IL-2-producing memory CD4+ T cells. (Sathaliyawala et al., 2013) and they tend to accumulate in mucosal tissues, and they outnumber CD8+ memory T cells in the mucosa. (Schreiner & King, 2018) Conventional TCRαβ+ CD4+d TCRαβ+ are found in the small intestine at 15–40%. The prevalence of conventional CD4+ T cells in the colon is 8–11 %. (Lutter et al., 2018)

In barrier immunity, Th17 cells play a prominent role. This effector T cell population is essential for microbial clearance, but it can also promote autoimmunity at barrier sites. (Marks & Craft, 2009). Even in the absence of an infection, effector CD4+ T cells in the lamina propria may secrete pro-inflammatory cytokines such as IL-17 and IFN-gamma. (Dubin & Kolls, 2008;

Sakaguchi et al., 2008; Schreiner & King, 2018). IL-17 production is confined to the intestinal mucosa. Nevertheless, the presence of IL-10-producing Treg cells could suppress this immune response. (Lutter et al., 2018; Sun et al., 2015)

Tregs have also been shown to play a role in maintaining immune homeostasis in the gut.(Sakaguchi et al., 2008; Sun et al., 2015). Thymus-derived CD4+ T cells present at the SLOs and they migrate to the intestinal epithelium by the gut homing signals. (Lutter et al., 2018) Treg requires β7 integrin for the migration to the gut by binding to MADCAM1 on gut venules. CCR7 relocates tissue residing Treg to the draining lymph nodes. (Pabst & Bernhardt, 2013) Additionally, Tregs are influenced by the gut microbiota. TCRαβ+CD4+ T cells' effector functions are curtailed, whereas Treg functions are enhanced. (Lutter et al., 2018)

IL-6 could also mediate the inflammatory response. IL-6 in synergy with TGF-β can induce RORγt expression and Th17 cell differentiation from naïve T cells. However, the presence of IL-6 could prevent TGF-β induced Foxp3 expression. TGF-β alone activates the Smad pathway, contributing to the downregulation of immune responses; IL-6 alone activates STAT3 transiently; however, when TGF- and IL-6 were combined, they induced a distinct transcriptional program. (Korn et al., 2009; Sun et al., 2015).

CD4+ T cells are predominantly found in the lamina propria; in particular, regulatory T (Treg) cells are relatively far more abundant there. (Lutter et al., 2018) When $Foxp3^+$ Tregs in the lamina propria migrate to the intestinal epithelium, they lose Foxp3 and convert to $Foxp3^-$ $CD8\alpha\alpha^+CD4^+$ T cells depending on the microbiota, which has an anti-inflammatory function. (Cheroutre et al., 2011) TCRαβ+CD4+ IELs could also express CD8αα homodimer, as a negative regulator of T cell activation. In support of this concept, Th2 $CD8^+\alpha\alpha^+CD4^+$ IELs, also gives $CD4^+TCR\alpha\beta$ IELs the ability to participate in intestinal immune tolerance consistent with Th cells' plasticity. (Lutter et al., 2018). In addition to the conventional CD4 T cell subset, $CD4^+TCR\alpha\beta^+$ IELs have T helper characteristics, $CD4^+TCR\alpha\beta^+$ IELs account for approximately 10–15% of the total IELs in humans. This correlation is attributed to the indispensable role of $CD4^+TCR\alpha\beta^+$ IELs in protective immunity. (Ma et al., 2021)

The expression of $CD8\alpha\alpha$ and the limited TCR repertoire support such tolerance, but it is insufficient. The intestinal microenvironment has been shown to reduce $CD4^+TCR\alpha\beta^+$ IELs' high reactivity. (Cheroutre et al., 2011; Ma et al., 2021)

CD4+ T cells are involved in the pathogenesis of celiac disease

The deamidated gliadin peptides can be presented to the gluten-specific $CD4^+$ T cells when activated in the context of the celiac disease-associated HLA molecules. Those peptides bind better to HLA- DQ2 and DQ8 molecules than their native counterparts. Gluten-specific CD4+ T cells secrete IFNγ and interleukin IL-21 after $CD4^+TCR\alpha\beta^+$ IELs mediate inflammation. (Ma et al., 2021).

CeD pathology is driven by gluten-reactive CD4+ T cells, which interact with B cells and CD8+ T cells. (Jabri and Sollid, 2017).CD4+ T cells are also critical for CD8+ IELs to become essential

T cells in the intestine, predominantly responsible for killing enterocytes. The local inflammatory response is more dependent on lamina propria cells, such as TCRαβ+CD4+ Th subsets. Moreover, conventional CD4+ IELs may also contribute to epithelial damage by upregulating natural killer cells. (Lutter et al., 2018) Gluten-specific CD4+T cells produce IFN-gamma, a characteristic of Th1 cells, along with other cytokines such as IL-4, IL-5, IL-10, TNF, and TGFβ. These findings are likely to present that Th1 immunity is developed against gluten and IL-10 production is produced by Th1 cell as a negative feedback loop. Additionally, overexpressed IL-15 in lamina propria enhances dendritic cells' function. (Depaolo et al., 2011) Upregulation of IL-15 drives Th1 immune responses against dietary antigens without systemic immunization. Type 1 IFN is also thought to link with IL-15 inflammatory T cell responses. (Monteleone et al., 2001).

The generation and activation of regulatory T cells (CD4 + CD25 + FOXP3 + Tregs) maintains tolerance to dietary proteins. Replacement of immunological tolerance by T cell-mediated hypersensitivity in celiac disease patients results in small intestinal damage and digestive symptoms. (Sollid and Jabri, 2013). Another link between CD4+ T cells and CeD pathogenesis is the dysfunction of Treg.

During the early development of immune cells, negative selection prevents T cells from reacting against self-antigens of the body tissues. (Klein et al., 2014). To display tolerance to food antigens, T cells should be limited in inflammatory responses to resident commensal microbes and foreign antigens by the virtue of immunological tolerance. Not all foreign antigens stimulate an excessive immune response. (Ma et al., 2021) Anergy, deletion of antigen-specific T cells and Treg differentiation in the thymus work together to build immune tolerance to self-antigens. (Apert et al., 2018; Saurer & Mueller, 2009)

Breakdown of oral immunity to gluten plays a crucial role in the formation of celiac disease. In this context, regulatory T cells elicit their role in preventing celiac disease by suppressing the anti-gluten response. $FOXP3^{+}$ T-reg cell, together with Type 1 (Tr1) regulatory T cells, are associated with IL-10 and TGF-β secretion. (Roncarolo et al. 2006; Sakaguchi et al. 2008). In celiac disease patients, DQ2.5-restricts Tr1 (Gianfrani et al. 2006). Under this model, HLA-DQ allotypes prevent regulatory cells from functioning in celiac disease patients. According to Cook, circulating FOXP3+CD39+ Tregs that become activated with gluten are also limited by DQ2.5. (Cook et al. 2017;Jabri and Sollid, 2017). However, most DQ2.5 subjects never develop celiac disease. There is no evidence for gluten-specific and DQ2.5-restricted regulatory T cells specificity for immunodominant gluten in healthy subjects (Christophersen et al. 2016 ; Jabri and Sollid, 2017).

According to one model, autoreactive TG2 antibodies play a specific role for CD4+ T cells. TG2s can form complex with gluten peptides and shows those peptides to specific CD4 T cells. Based on the samples obtained by the healthy controls and patients' blood, healthy subjects demonstrate they are lack such peptide specific memory and regulatory T cells. On the other hand, gut biopsies from the CeD patients have gluten-specific CD4+ T cells at a frequency of 0.1-1.2%. (Bodd et al., 2013).

Putative treatments targeting CD4+ T cells

The majority of the autoimmune disease is caused by the imbalance in Treg cells and T effector cells (Teff). Still, the underlying mechanism for losing Treg remained elusive: either Treg suppression is blocked by Teff and results in celiac disease, or Treg cell expansion becomes insufficient. The modulation of the balance of T cells for the therapeutic approach to celiac disease has moved from the basis of effector T cells resisting to Treg cell suppression to the basis of amplifying the Treg cells. Adherence to a gluten-free diet (GFD) is a current solution to villous abnormalities. It decreases the frequencies of gluten-specific $TCRab^+$ $CD4^+$ T cells in the lamina propria and cytolytic $TCRab^+$ $CD8^+$ IELs, which are the main contributor to CeD pathogenesis (Jabri and Sollid, 2009).

Additionally, we could suggest a model of 'tuned suppression' for promoting Treg upon encountering inflammatory stimuli. Modulation in TLR ligation could also suppress the immune response, which constitutes activation of naïve T cells through Toll-like receptor-ligand binding on membrane molecules and the production of cytokines and chemokines. The excessive immune response can be effectively curtailed as soon as effector cells reacquire suppression sensitivity. Additionally, FoxP3 expression could be enhanced due to its critical role in suppressive function, and deficiency of the Foxp3 gene leads to autoimmune diseases. (Apert et al., 2018; Sun et al., 2015).

An alternative treatment that is implemented in IBD treatment could be blocking α4β7. It could lead to less-gut-specific homing markers to ensure to prevent lymphocyte trafficking to the gut.(Lutter et al., 2018). Such blocking antibodies already exist and are used in treatment of inflammatory bowel disease. However, such approaches may not work if the disease is driven by local resident cell populations. In the next section we will discuss the development of local tissue resident memory.

Memory T cells and their phenotypic markers

Table 2. Representation of direct and inderect relationship of selected CD marker expressions to CD8 TEM &TRM, CD4 TEM &TRM

	CD28	CD127	CD69	CD103	CCR7	CD62L	CD45RO
Naïve T cell	+	+	+	-	+	-	-
CD8 TEM in lymphoid tissue	+	-	+	-	+/-	-	+
CD8 TRM in the mucosa	-	+	+	+	-	+	+
CD4 TEM	+/-	-	-	-	-	-	+
CD4 TRM	+/-	-	+	+/-	-	-	+

CD8+T

Many CD8+ T Cells in the intestine display TRM marker expression, CD103, and CD69 (Bergsbaken et al., 2017) (Table2). CXCR3 is a significant marker that enables recruiting CD8+ T cells to pro-inflammatory microenvironments, hence makes them differentiate into the CD103+ TRM population. (Bergsbaken & Bevan, 2015). An important cytokine that drives CD8+ T cells to differentiate into CD103+ TRM is TGF-β. TGFβ induces downregulation of T-bet, which gives rise to differentiation of tissue-resident CD8 + T cell formation. (Samat et al., 2021). In contrast, pro-inflammatory cytokines, such as type I interferons and IL-12, move TRM to CD103– TRM population (Bergsbaken et al., 2017; Seder et al., 2008).

Recent studies with single-cell RNA sequencing demonstrated two populations of CD8+ T cells in the intestine. One of them expresses ITGAE (CD103) and CD7, IL7R (CD127), in which IL7 is the cytokine that provides a homeostatic proliferation of lymphocytes. (Raeber et al., 2018). It is also a survival factor for epidermal TRM cell persistence. (FitzPatrick et al., 2021).
CD161 + CD8+ T cell population is another population that is found in the intestine. (Kondo et al., 2007) They secrete IL-22, a cytokine involved in tissue repair and epithelial defence. (Billerbeck et al., 2010). CD161, C type lectin-like receptor, is identified as its feature for promoting antigen-dependent T cell proliferation. CCR6+ CD8+ T cells express a high level of granzyme A and B. Still, the perforin level in this subset is low. (Masopust et al., 2010)

CD4+T

The other population expresses the integrin ITGB2 (CD18, β 2-integrin), expresses low levels of ITGAE(CD103) but high levels of KLRG1. Therefore, small intestinal CD103+ $CD8^+$ T could also be differentiated based on the expression of surface KLRG1.(Bartolomé-Casado et al., 2019; FitzPatrick et al., 2021). The population that expresses β 2-integrin and CD18 also express cytotoxic receptors and granzymes. Those transcriptionally distinct subsets display differences in terms of function and localization. (FitzPatrick et al., 2021).

Foxp3 is a unique transcription factor that distinguishes the Treg cell subset, and CD25 is a surface marker for Treg cells. (Pabst & Bernhardt, 2013; Sakaguchi et al., 2008; Sun et al., 2015) Tregs are generally produced in the thymus through IL-2 signaling. There is another subset of Tregs, which is developed from Foxp3+ CD25+ CD4+ cells in peripheral tissues. Additionally, CD127 expression inversely correlates with FoxP3 expression.

CD127 is downregulated on CD4 T cells on peripheral tissues. (W. Liu et al., 2006) TGF-β and IL-2 induce expression of FoxP3 of activated CD4+ T cells. (Pabst & Bernhardt, 2013) FoxP3 facilitates the production of pro-inflammatory cytokines. Therefore, FoxP3 and TGF-β expressing cells are generally present at the site of inflammation and suppress that inflammation. (Lutter et al., 2018; O'Shea & Paul, 2010).

Discussion

CeD is an autoimmune disease driven by the aberrant response to the gluten proteins (Jabri and Sollid, 2017). This response is led by a range of T-cell subsets that have distinct but crucial roles in the immunopathology of the disease (Jabri et al. 2000). Of these subsets, CD4+ T-cells mediate its affects via activating CD8+ T-cells and B-cells (Jabri and Sollid, 2017). Furthermore, they have a role in enhancing the antigen presenting ability of dentritic cells. (Depaolo et al., 2011). CD4+ T-cells also secrete Th1-related cytokines such as IFNγ, which further shifts the balance to Th1-response and support inflammation in the small intestines (Dubin & Kolls, 2008; Sakaguchi et al., 2008; Schreiner & King, 2018). On the other hand, CD8+ IELs inflict injury to intestine tissue and cause villous atrophy by targeting distressed intestinal epithelial cells (Konjar et al., 2017; Mazzarella et al., 2008). CD8+ IELs of CeD patients have especially low threshold for activation due to high levels of IL-15 in these patients, which makes more reactive to stimuli and increases self-injury (Halstensen & Brandtzaeg, 1993; Samat et al., 2021). At the same time, they are also another source of IFNγ and other Th1 cytokines (Mazzarella et al., 2008). Furthermore, CD8+ IELs attract other immune cells by chemokines such as CCR9 and RGS1 (Samat et al., 2021). Both CD4+ and CD8+ T-cells have a TRM phenotype and cells with this phenotype are critical in the immunopathogenesis of CeD. CD8+ TRMs play an especially important role in CeD as these cells have higher cytotoxic capabilities against epithelial cells, and they can be activated even without the presence of gluten proteins via an TCR-independent mechanism. Additionally, they cans also suppress the influx of circulating T cells, further causing an imbalance in the T-cell population in the gut (Hayday et al., 2001; Kutlu et al., 1993).

One of the major source of the immunopathology of CeD is the lack of sensitivity of the mentioned T-cell subsets to Tregs, which serve as the brakes against excessive inflammation (Sollid and Jabri, 2013). Novel therapeutical approaches could be developed to overcome this lack of T-reg sensitivity. As CD8+ IELs are involved in the inhibition of Treg function, such approaches may target them. An example would be blocking IL-15, a cytokine critical for the activation of CD8+ IELs in CeD (Abadie & Jabri, 2014; Cheroutre et al., 2011). IFN-γ could also be targeted as it is also one of the major drivers of CeD immunopathology (Schreiner & King, 2018). A different approach to dampen CD8+ IEL-mediated tissue injury could be preventing IELs migration to small intestines by blocking the gut-homing α4β7 integrin, using drugs such as Vedolizumab (Samat et al., 2021). These cells could also be indirectly targeted by blocking another cytokine, IL-2, which plays a role in their memory cell expansion (Apert et al., 2018). Furthermore, to mitigate the aberrant pro-inflammatory cytokine activity could be administering anti-inflammatory cytokines to CeD patients. IL-10 is a promising candidate for this purpose due to its established suppressive effects on T-cells (Forsberg et al., 2007).

Conclusion

The aim was to understand the memory T-cells and their function within CeD. Immunopathogenesis of CeD is visually discussed and the importance of CD8+T and CD4+T reiterated. These two memory cells are then evaluated with their respective IELs and Tregs with suggested putative treatment techniques. The further consideration of phenotypic markers enabled deeper understanding on the subject matter. Certain ILs, and especially IL15

– as it is crucial for long term maintenance of TRMs and blockade of IL15s, using monoclonal antibodies could potentially be treating autoimmune disorders – are further included as of several cytokines and chemokines are correlated with celiac disease.

References

Abadie, V., & Jabri, B. (2014). IL-15: A central regulator of celiac disease immunopathology. In *Immunological Reviews* (Vol. 260, Issue 1). https://doi.org/10.1111/imr.12191

Annunziato, F., & Romagnani, S. (2009). Heterogeneity of human effector CD4+ +T cells. In *Arthritis Research and Therapy* (Vol. 11, Issue 6). https://doi.org/10.1186/ar2843

Apert, C., Romagnoli, P., & van Meerwijk, J. P. M. (2018). IL-2 and IL-15 dependent thymic development of Foxp3-expressing regulatory T lymphocytes. In *Protein and Cell* (Vol. 9, Issue 4). https://doi.org/10.1007/s13238-017-0425-3

Bartolomé-Casado, R., Landsverk, O. J. B., Chauhan, S. K., Richter, L., Phung, D., Greiff, V., Risnes, L. F., Yao, Y., Neumann, R. S., Yaqub, S., Øyen, O., Horneland, R., Aandahl, E. M., Paulsen, V., Sollid, L. M., Qiao, S. W., Baekkevold, E. S., & Jahnsen, F. L. (2019). Resident memory CD8 T cells persist for years in human small intestine. *Journal of Experimental Medicine*, *216*(10). https://doi.org/10.1084/jem.20190414

Bergsbaken, T., & Bevan, M. J. (2015). Proinflammatory microenvironments within the intestine regulate the differentiation of tissue-resident CD8+ T cells responding to infection. *Nature Immunology*, *16*(4). https://doi.org/10.1038/ni.3108

Bergsbaken, T., Bevan, M. J., & Fink, P. J. (2017). Local Inflammatory Cues Regulate Differentiation and Persistence of CD8+ Tissue-Resident Memory T Cells. *Cell Reports*, *19*(1). https://doi.org/10.1016/j.celrep.2017.03.031

Billerbeck, E., Kang, Y. H., Walker, L., Lockstone, H., Grafmueller, S., Fleming, V., Flint, J., Willberg, C. B., Bengsch, B., Seigel, B., Ramamurthy, N., Zitzmann, N., Barnes, E. J., Thevanayagam, J., Bhagwanani, A., Leslie, A., Oo, Y. H., Kollnberger, S., Bowness, P., … Klenerman, P. (2010). Analysis of CD161 expression on human CD8+ T cells defines a distinct functional subset with tissue-homing properties. *Proceedings of the National Academy of Sciences of the United States of America*, *107*(7). https://doi.org/10.1073/pnas.0914839107

Bodd, M., Ráki, M., Bergseng, E., Jahnsen, J., Lundin, K. E. A., & Sollid, L. M. (2013). Direct cloning and tetramer staining to measure the frequency of intestinal gluten-reactive T cells in celiac disease. *European Journal of Immunology*, *43*(10). https://doi.org/10.1002/eji.201343382

Bottois, H., Ngollo, M., Hammoudi, N., Courau, T., Bonnereau, J., Chardiny, V., Grand, C., Gergaud, B., Allez, M., & Le Bourhis, L. (2020). KLRG1 and CD103 Expressions Define Distinct Intestinal Tissue-Resident Memory CD8 T Cell Subsets Modulated in Crohn's Disease. *Frontiers in Immunology*, *11*. https://doi.org/10.3389/fimmu.2020.00896

Boyman, O., & Sprent, J. (2012). The role of interleukin-2 during homeostasis and activation of the immune system. In *Nature Reviews Immunology* (Vol. 12, Issue 3). https://doi.org/10.1038/nri3156

Casey, K. A., Fraser, K. A., Schenkel, J. M., Moran, A., Abt, M. C., Beura, L. K., Lucas, P. J., Artis, D., Wherry, E. J., Hogquist, K., Vezys, V., & Masopust, D. (2012). Antigen-Independent Differentiation and Maintenance of Effector-like Resident Memory T Cells in Tissues. *The Journal of Immunology*, *188*(10). https://doi.org/10.4049/jimmunol.1200402

Cheroutre, H., Lambolez, F., & Mucida, D. (2011). The light and dark sides of intestinal intraepithelial lymphocytes. In *Nature Reviews Immunology* (Vol. 11, Issue 7). https://doi.org/10.1038/nri3007

Cibrián, D., & Sánchez-Madrid, F. (2017). CD69: from activation marker to metabolic

gatekeeper. In *European Journal of Immunology* (Vol. 47, Issue 6). https://doi.org/10.1002/eji.201646837

de Leur, K., Dieterich, M., Hesselink, D. A., Corneth, O. B. J., Dor, F. J. M. F., de Graav, G. N., Peeters, A. M. A., Mulder, A., Kimenai, H. J. A. N., Claas, F. H. J., Clahsen-van Groningen, M. C., van der Laan, L. J. W., Hendriks, R. W., & Baan, C. C. (2019). Characterization of donor and recipient CD8+ tissue-resident memory T cells in transplant nephrectomies. *Scientific Reports*, *9*(1). https://doi.org/10.1038/s41598-019-42401-9

Debes, G. F., Arnold, C. N., Young, A. J., Krautwald, S., Lipp, M., Hay, J. B., & Butcher, E. C. (2005). Chemokine receptor CCR7 required for T lymphocyte exit from peripheral tissues. *Nature Immunology*, *6*(9). https://doi.org/10.1038/ni1238

Depaolo, R. W., Abadie, V., Tang, F., Fehlner-Peach, H., Hall, J. A., Wang, W., Marietta, E. V., Kasarda, D. D., Waldmann, T. A., Murray, J. A., Semrad, C., Kupfer, S. S., Belkaid, Y., Guandalini, S., & Jabri, B. (2011). Co-adjuvant effects of retinoic acid and IL-15 induce inflammatory immunity to dietary antigens. *Nature*, *471*(7337). https://doi.org/10.1038/nature09849

Dubin, P. J., & Kolls, J. K. (2008). Th17 cytokines and mucosal immunity. In *Immunological Reviews* (Vol. 226, Issue 1). https://doi.org/10.1111/j.1600-065X.2008.00703.x

FitzPatrick, M. E. B., Provine, N. M., Garner, L. C., Powell, K., Amini, A., Irwin, S. L., Ferry, H., Ambrose, T., Friend, P., Vrakas, G., Reddy, S., Soilleux, E., Klenerman, P., & Allan, P. J. (2021). Human intestinal tissue-resident memory T cells comprise transcriptionally and functionally distinct subsets. *Cell Reports*, *34*(3). https://doi.org/10.1016/j.celrep.2020.108661

Forsberg, G., Hernell, O., Hammarström, S., & Hammarström, M. L. (2007). Concomitant increase of IL-10 and pro-inflammatory cytokines in intraepithelial lymphocyte subsets in celiac disease. *International Immunology*, *19*(8). https://doi.org/10.1093/intimm/dxm077

Golubovskaya, V., & Wu, L. (2016). Different subsets of T cells, memory, effector functions, and CAR-T immunotherapy. In *Cancers* (Vol. 8, Issue 3). MDPI AG. https://doi.org/10.3390/cancers8030036

Halstensen, T. S., & Brandtzaeg, P. (1993). Activated T lymphocytes in the celiac lesion: Non-proliferative activation (CD25) of CD4+ α/β cells in the lamina propria but proliferation (Ki-67) of α/β and γ/δ cells in the epithelium. *European Journal of Immunology*, *23*(2). https://doi.org/10.1002/eji.1830230231

Iijima, N., & Iwasaki, A. (2015). Tissue instruction for migration and retention of TRM cells. In *Trends in Immunology* (Vol. 36, Issue 9). https://doi.org/10.1016/j.it.2015.07.002

Jabri, B., & Abadie, V. (2015). IL-15 functions as a danger signal to regulate tissue-resident T cells and tissue destruction. In *Nature Reviews Immunology* (Vol. 15, Issue 12). https://doi.org/10.1038/nri3919

Kanno, Y., Vahedi, G., Hirahara, K., Singleton, K., & O'Shea, J. J. (2012). Transcriptional and epigenetic control of T helper cell specification: Molecular mechanisms underlying commitment and plasticity. In *Annual Review of Immunology* (Vol. 30). https://doi.org/10.1146/annurev-immunol-020711-075058

Khan, U., & Ghazanfar, H. (2018). T Lymphocytes and Autoimmunity. *International Review of Cell and Molecular Biology*, *341*. https://doi.org/10.1016/bs.ircmb.2018.05.008

Klein, L., Kyewski, B., Allen, P. M., & Hogquist, K. A. (2014). Positive and negative selection of the T cell repertoire: What thymocytes see (and don't see). In *Nature Reviews Immunology* (Vol. 14, Issue 6, pp. 377–391). Nature Publishing Group.

https://doi.org/10.1038/nri3667
Kondo, T., Takata, H., & Takiguchi, M. (2007). Functional expression of chemokine receptor CCR6 on human effector memory CD8+ T cells. *European Journal of Immunology*, *37*(1). https://doi.org/10.1002/eji.200636251
Konjar, Š., Ferreira, C., Blankenhaus, B., & Veldhoen, M. (2017). Intestinal barrier interactions with specialized CD8 T cells. In *Frontiers in Immunology* (Vol. 8, Issue OCT). https://doi.org/10.3389/fimmu.2017.01281
Kool, M., Hammad, H., & Lambrecht, B. N. (2012). Cellular networks controlling Th2 polarization in allergy and immunity. In *F1000 Biology Reports* (Vol. 4, Issue 1). https://doi.org/10.3410/B4-6
Korn, T., Bettelli, E., Oukka, M., & Kuchroo, V. K. (2009). IL-17 and Th17 Cells. *Annual Review of Immunology*, *27*(1), 485–517. https://doi.org/10.1146/annurev.immunol.021908.132710
Kumar, B. V., Ma, W., Miron, M., Granot, T., Guyer, R. S., Carpenter, D. J., Senda, T., Sun, X., Ho, S. H., Lerner, H., Friedman, A. L., Shen, Y., & Farber, D. L. (2017). Human Tissue-Resident Memory T Cells Are Defined by Core Transcriptional and Functional Signatures in Lymphoid and Mucosal Sites. *Cell Reports*, *20*(12). https://doi.org/10.1016/j.celrep.2017.08.078
Kunkel, E. J., Campbell, J. J., Haraldsen, G., Pan, J., Boisvert, J., Roberts, A. I., Ebert, E. C., Vierra, M. A., Goodman, S. B., Genovese, M. C., Wardlaw, A. J., Greenberg, H. B., Parker, C. M., Butcher, E. C., Andrew, D. P., & Agace, W. W. (2000). Lymphocyte CC chemokine receptor 9 and epithelial thymus-expressed chemokine (TECK) expression distinguish the small intestinal immune compartment: Epithelial expression of tissue-specific chemokines as an organizing principle in regional immunity. *Journal of Experimental Medicine*, *192*(5). https://doi.org/10.1084/jem.192.5.761
Liu, W., Putnam, A. L., Xu-yu, Z., Szot, G. L., Lee, M. R., Zhu, S., Gottlieb, P. A., Kapranov, P., Gingeras, T. R., Barbara, B. F., Clayberger, C., Soper, D. M., Ziegler, S. F., & Bluestone, J. A. (2006). CD127 expression inversely correlates with FoxP3 and suppressive function of human CD4+ T reg cells. *Journal of Experimental Medicine*, *203*(7), 1701–1711. https://doi.org/10.1084/jem.20060772
Liu, Y., Ma, C., & Zhang, N. (2018). Tissue-specific control of tissue-resident memory T cells. *Critical Reviews in Immunology*, *38*(2). https://doi.org/10.1615/CritRevImmunol.2018025653
Lutter, L., Hoytema van Konijnenburg, D. P., Brand, E. C., Oldenburg, B., & van Wijk, F. (2018). The elusive case of human intraepithelial T cells in gut homeostasis and inflammation. In *Nature Reviews Gastroenterology and Hepatology* (Vol. 15, Issue 10). https://doi.org/10.1038/s41575-018-0039-0
Ma, H., Qiu, Y., & Yang, H. (2021). Intestinal intraepithelial lymphocytes: Maintainers of intestinal immune tolerance and regulators of intestinal immunity. In *Journal of Leukocyte Biology* (Vol. 109, Issue 2). https://doi.org/10.1002/JLB.3RU0220-111
Marks, B. R., & Craft, J. (2009). Barrier immunity and IL-17. In *Seminars in Immunology* (Vol. 21, Issue 3). https://doi.org/10.1016/j.smim.2009.03.001
Masopust, D., Choo, D., Vezys, V., Wherry, E. J., Duraiswamy, J., Akondy, R., Wang, J., Casey, K. A., Barber, D. L., Kawamura, K. S., Fraser, K. A., Webby, R. J., Brinkmann, V., Butcher, E. C., Newell, K. A., & Ahmed, R. (2010). Dynamic T cell migration program provides resident memory within intestinal epithelium. *Journal of Experimental Medicine*, *207*(3). https://doi.org/10.1084/jem.20090858

Mazzarella, G., Stefanile, R., Camarca, A., Giliberti, P., Cosentini, E., Marano, C., Iaquinto, G., Giardullo, N., Auricchio, S., Sette, A., Troncone, R., & Gianfrani, C. (2008). Gliadin Activates HLA Class I-Restricted CD8+ T Cells in Celiac Disease Intestinal Mucosa and Induces the Enterocyte Apoptosis. *Gastroenterology*, *134*(4). https://doi.org/10.1053/j.gastro.2008.01.008

Mueller, S. N., & Mackay, L. K. (2016). Tissue-resident memory T cells: Local specialists in immune defence. In *Nature Reviews Immunology* (Vol. 16, Issue 2). https://doi.org/10.1038/nri.2015.3

O'Shea, J., & Paul, W. E. (2010). Mechanisms underlying lineage commitment and plasticity of helper CD4 + T cells. In *Science* (Vol. 327, Issue 5969). https://doi.org/10.1126/science.1178334

Pabst, O., & Bernhardt, G. (2013). On the road to tolerance - Generation and migration of gut regulatory T cells. *European Journal of Immunology*, *43*(6). https://doi.org/10.1002/eji.201243154

Park, C. O., & Kupper, T. S. (2015). The emerging role of resident memory T cells in protective immunity and inflammatory disease. In *Nature Medicine* (Vol. 21, Issue 7). https://doi.org/10.1038/nm.3883

Pender, M. P. (2012). CD8+ t-cell deficiency, epstein-barr virus infection, vitamin d deficiency, and steps to autoimmunity: A unifying hypothesis. In *Autoimmune Diseases* (Vol. 1, Issue 1). https://doi.org/10.1155/2012/189096

Pennock, N. D., White, J. T., Cross, E. W., Cheney, E. E., Tamburini, B. A., & Kedl, R. M. (2013). T cell responses: Naïve to memory and everything in between. *American Journal of Physiology - Advances in Physiology Education*, *37*(4). https://doi.org/10.1152/advan.00066.2013

Raeber, M. E., Zurbuchen, Y., Impellizzieri, D., & Boyman, O. (2018). The role of cytokines in T-cell memory in health and disease. In *Immunological Reviews* (Vol. 283, Issue 1). https://doi.org/10.1111/imr.12644

Sakaguchi, S., Yamaguchi, T., Nomura, T., & Ono, M. (2008). Regulatory T Cells and Immune Tolerance. In *Cell* (Vol. 133, Issue 5). https://doi.org/10.1016/j.cell.2008.05.009

Samat, A. A. K., van der Geest, J., Vastert, S. J., van Loosdregt, J., & van Wijk, F. (2021). Tissue-Resident Memory T Cells in Chronic Inflammation-Local Cells with Systemic Effects? In *Cells* (Vol. 10, Issue 2). https://doi.org/10.3390/cells10020409

Sánchez-Castañon, M., Castro, B. G., Toca, M., Santacruz, C., Arias-Loste, M., Iruzubieta, P., Crespo, J., & López-Hoyos, M. (2016). Intraepithelial lymphocytes subsets in different forms of celiac disease. *Autoimmunity Highlights*, *7*(1). https://doi.org/10.1007/s13317-016-0085-y

Saraiva, M., & O'Garra, A. (2010). The regulation of IL-10 production by immune cells. In *Nature Reviews Immunology* (Vol. 10, Issue 3). https://doi.org/10.1038/nri2711

Sasson, S. C., Gordon, C. L., Christo, S. N., Klenerman, P., & Mackay, L. K. (2020). Local heroes or villains: tissue-resident memory T cells in human health and disease. In *Cellular and Molecular Immunology* (Vol. 17, Issue 2). https://doi.org/10.1038/s41423-019-0359-1

Sathaliyawala, T., Kubota, M., Yudanin, N., Turner, D., Camp, P., Thome, J. J. C., Bickham, K. L., Lerner, H., Goldstein, M., Sykes, M., Kato, T., & Farber, D. L. (2013). Distribution and Compartmentalization of Human Circulating and Tissue-Resident Memory T Cell Subsets. *Immunity*, *38*(1), 187–197. https://doi.org/10.1016/j.immuni.2012.09.020

Saurer, L., & Mueller, C. (2009). T cell-mediated immunoregulation in the gastrointestinal tract. In *Allergy: European Journal of Allergy and Clinical Immunology* (Vol. 64, Issue 4).

https://doi.org/10.1111/j.1398-9995.2009.01965.x
Schenkel, J. M., Fraser, K. A., Beura, L. K., Pauken, K. E., Vezys, V., & Masopust, D. (2014). Resident memory CD8 t cells trigger protective innate and adaptive immune responses. *Science*, *346*(6205). https://doi.org/10.1126/science.1254536
Schreiner, D., & King, C. G. (2018). CD4+ memory T cells at home in the tissue: Mechanisms for health and disease. In *Frontiers in Immunology* (Vol. 9, Issue OCT). https://doi.org/10.3389/fimmu.2018.02394
Seder, R. A., & Ahmed, R. (2003). Similarities and differences in CD4+ and CD8+ effector and memory T cell generation. In *Nature Immunology* (Vol. 4, Issue 9). https://doi.org/10.1038/ni969
Seder, R. A., Darrah, P. A., & Roederer, M. (2008). T-cell quality in memory and protection: Implications for vaccine design. In *Nature Reviews Immunology* (Vol. 8, Issue 4). https://doi.org/10.1038/nri2274
Shale, M., Schiering, C., & Powrie, F. (2013). CD4+ T-cell subsets in intestinal inflammation. *Immunological Reviews*, *252*(1). https://doi.org/10.1111/imr.12039
Sheridan, B. S., & Lefrançois, L. (2010). Intraepithelial lymphocytes: To serve and protect. In *Current Gastroenterology Reports* (Vol. 12, Issue 6). https://doi.org/10.1007/s11894-010-0148-6
Sheridan, B. S., & Lefrançois, L. (2011). Regional and mucosal memory T cells. In *Nature Immunology* (Vol. 12, Issue 6). https://doi.org/10.1038/ni.2029
Sheridan, B. S., Pham, Q. M., Lee, Y. T., Cauley, L. S., Puddington, L., & Lefrançois, L. (2014). Oral infection drives a distinct population of intestinal resident memory cd8+ t cells with enhanced protective function. *Immunity*, *40*(5). https://doi.org/10.1016/j.immuni.2014.03.007
Skapenko, A., Leipe, J., Lipsky, P. E., & Schulze-Koops, H. (2005). The role of the T cell in autoimmune inflammation. In *Arthritis Research and Therapy* (Vol. 7, Issue SUPPL. 2). https://doi.org/10.1186/ar1505
Sollid, L. M. (2004). Intraepithelial lymphocytes in celiac disease: License to kill revealed. In *Immunity* (Vol. 21, Issue 3). https://doi.org/10.1016/j.immuni.2004.08.002
Steinbach, K., Vincenti, I., & Merkler, D. (2018). Resident-Memory T Cells in tissue-restricted immune responses: For better or worse? In *Frontiers in Immunology* (Vol. 9, Issue NOV). https://doi.org/10.3389/fimmu.2018.02827
Sun, M., He, C., Cong, Y., & Liu, Z. (2015). Regulatory immune cells in regulation of intestinal inflammatory response to microbiota. In *Mucosal Immunology* (Vol. 8, Issue 5). https://doi.org/10.1038/mi.2015.49
Tai, Y., Wang, Q., Korner, H., Zhang, L., & Wei, W. (2018). Molecular mechanisms of T cells activation by dendritic cells in autoimmune diseases. In *Frontiers in Pharmacology* (Vol. 9, Issue JUN). https://doi.org/10.3389/fphar.2018.00642
Topham, D. J., & Reilly, E. C. (2018). Tissue-resident memory CD8+ T cells: From phenotype to function. In *Frontiers in Immunology* (Vol. 9, Issue MAR). https://doi.org/10.3389/fimmu.2018.00515
Whiteside, S. K., Snook, J. P., Williams, M. A., & Weis, J. J. (2018). Bystander T Cells: A Balancing Act of Friends and Foes. In *Trends in Immunology* (Vol. 39, Issue 12). https://doi.org/10.1016/j.it.2018.10.003
Williams, M. A., Tyznik, A. J., & Bevan, M. J. (2006). Interleukin-2 signals during priming are required for secondary expansion of CD8+ memory T cells. *Nature*, *441*(7095). https://doi.org/10.1038/nature04790

Willinger, T., Freeman, T., Hasegawa, H., McMichael, A. J., & Callan, M. F. C. (2005). Molecular Signatures Distinguish Human Central Memory from Effector Memory CD8 T Cell Subsets. *The Journal of Immunology*, *175*(9), 5895–5903. https://doi.org/10.4049/jimmunol.175.9.5895

Winter, S. J., & Krueger, A. (2019). Development of unconventional t cells controlled by microrna. In *Frontiers in Immunology* (Vol. 10, Issue OCT). https://doi.org/10.3389/fimmu.2019.02520

Wu, H., Liao, W., Li, Q., Long, H., Yin, H., Zhao, M., Chan, V., Lau, C. S., & Lu, Q. (2018). Pathogenic role of tissue-resident memory T cells in autoimmune diseases. In *Autoimmunity Reviews* (Vol. 17, Issue 9). https://doi.org/10.1016/j.autrev.2018.03.014

Zhu, J. (2015). T helper 2 (Th2) cell differentiation, type 2 innate lymphoid cell (ILC2) development and regulation of interleukin-4 (IL-4) and IL-13 production. In *Cytokine* (Vol. 75, Issue 1). https://doi.org/10.1016/j.cyto.2015.05.010

Zhu, J., Yamane, H., & Paul, W. E. (2010). Differentiation of effector CD4+ T cell populations. In *Annual Review of Immunology* (Vol. 28). https://doi.org/10.1146/annurev-immunol-030409-101212