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Review Article

Investigating Human Liver Tissue-Resident Memory T Cells from the Perspectives of Gastroenterologists and Hepatologists

Ji Won Han[1,2], Eui-Cheol Shin[3,4]

[1]The Catholic University Liver Research Center, College of Medicine, The Catholic University of Korea, Seoul, Korea; [2]Department of Internal Medicine, Seoul St. Mary's Hospital, College of Medicine, The Catholic University of Korea, Seoul, Korea; [3]Graduate School of Medical Science and Engineering, Korea Advanced Institute of Science and Technology (KAIST), Daejeon, Korea; [4]The Center for Viral Immunology, Korea Virus Research Institute, Institute for Basic Science (IBS), Daejeon, Korea

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Corresponding Author

Ji Won Han
ORCID https://orcid.org/0000-0003-1456-1450
E-mail tmznjf@catholic.ac.kr.

Eui-Cheol Shin
ORCID https://orcid.org/0000-0002-6308-9503
E-mail ecshin@kaist.ac.kr.

Liver tissue-resident memory T (T_{RM}) cells play a pivotal role in hepatic immune responses. Their unique residence within liver sinusoids allow continuous antigen surveillance. In this review, we highlight the role of liver T_{RM} cells in protective immunity and disease pathology. Comparisons between human and murine liver T_{RM} cells reveal species-specific characteristics, suggesting the need for human-focused studies. One key finding is the involvement of liver T_{RM} cells in viral hepatitis, where they can both control infection and contribute to liver damage. Liver T_{RM} cells also exhibit dual roles in metabolic-associated steatotic liver disease, promoting inflammation and fibrosis while also contributing to fibrosis resolution. In autoimmune liver diseases, such as autoimmune hepatitis and primary sclerosing cholangitis, the presence of liver T_{RM} cells correlates with disease severity. In this review, we underscore the importance of liver T_{RM} cells in vaccine development, particularly vaccines against malaria. Future research should focus on the mechanisms governing T_{RM}-cell formation, maintenance, and function, with the aim of supporting their protective roles while mitigating detrimental effects. Advancing our understanding of liver T_{RM} cells will enhance our knowledge of liver immunology and inform novel therapeutic strategies for liver disease management. **(Gut Liver 2025;19:161-170)**

Key Words: T-lymphocytes; Tissue resident T cell; Liver diseases

INTRODUCTION

Most immune cells, including over 95% of T cells, reside and function in tissues rather than in blood.[1] These tissues include lymphoid organs (e.g., bone marrow, spleen, and lymph nodes) and barrier surfaces (e.g., the skin, gut, and mucous membranes). Key populations of tissue-resident immune cells include tissue-resident memory T (T_{RM}) cells, dendritic cells, macrophages, and innate lymphoid cells, which are crucial for local immune responses.[2] T cells constitute an adaptive immune cell population that plays a central role in immune defense. Notably, $CD8^+$ T cells eliminate infected, damaged, or tumor cells, while $CD4^+$ T cells facilitate and regulate immune responses. The most important feature of the adaptive immune system is the formation of memory T cells, which enable a rapid and effective response upon re-exposure to pathogens. T_{RM} cells might serve this role immediately at the entry sites of various pathogens.[3]

The liver is a vital immunological organ, buffering gut contents and systemic circulation. About 80% of the liver's blood supply comes from the gut via the portal vein. This blood is rich in dietary and microbial antigens, which must be processed by the liver as it performs immunosurveillance.[4] Additionally, the liver has a distinctive anatomical vascular system, which allows continuous connection between immune cells, liver sinusoid endothelial cells (LSEC), and hepatocytes. Notably, its low-pressure blood flow and fenestrated endothelium facilitate interactions between immune cells and hepatic cells.[5] The liver sinusoids and the space of Disse are populated by immune cells that maintain organ homeostasis and regulate inflammation. These

cells adapt to this unique environment, adopting unique characteristics compared to the circulating population. For example, liver $CD8^+$ T_{RM} cells exhibit special characteristics compared to circulating T cells, and play crucial roles in liver immunity, participating in both the initiation and resolution of intrahepatic inflammation. Dysregulation of T_{RM} cells is implicated in the pathogenesis of various liver diseases.[6]

In this review, we aim to provide valuable summaries for clinicians, such as gastroenterologists and hepatologists conducting clinical and translational research. We review the general features of $CD8^+$ T_{RM} cells, with specific focus on the characteristics of liver T_{RM} cells. We will also compare liver T_{RM} cells between humans and mice, which supports the necessity of investigating this population using human samples. Finally, we will summarize the clinical implications of the present knowledge of liver T_{RM} cells in human liver diseases.

GENERAL FEATURES OF T_{RM} CELLS

1. Phenotypic and transcriptional characteristics of T_{RM} cells

The term T_{RM} cells generally refers to conventional $CD8^+$ memory (not naïve) T cells with αβ T cell receptor characterized by the expression of CD69 and/or CD103, which are not expressed in circulating T cells.[7] CD69 and CD103 help T_{RM} cells reside in peripheral tissues, such as the epithelium, through interaction with molecules like E-cadherin and by inhibiting *KLF2* and *S1PR1*, which facilitate egress from peripheral organs.[8] T_{RM} cells also generally express CD49a and CXCR6,[9] and lack markers typically found in central homing memory T cells, such as CCR7 and CD62L.[10] One key transcription factor is *Runx3*, which is crucial for T_{RM}-cell differentiation.[11] Additionally, *Hobit* and *Blimp* are involved in driving T_{RM}-cell differentiation, and have been described as important transcriptional regulators.[12] Epigenetic modifications, like DNA methylation and histone acetylation, might also regulate T_{RM}-cell development and function. DNA methylation is reduced in key genes such as *PRF1, CD39,* and *CD103*, while histone acetylation support expressions of these genes in T_{RM} cells.[13,14] These findings indicate that T_{RM} cells have distinct phenotypic characteristics compared to circulating T cells (Fig. 1).

2. Generation and maintenance of T_{RM} cells

Multiple exposures to antigens can generate large pools of T_{RM} cells without altering the pre-existing T-cell pool, suggesting that antigen stimulation plays an important role in T_{RM}-cell generation.[15] Some reports indicate that antigen stimulation is also required to maintain this population.[16] Additionally, cytokines such as interleukin (IL)-15 have been considered critical for T_{RM}-cell differentiation and survival, through mechanisms involving mTOR signaling pathways.[17-20] Transforming growth factor (TGF)-β is also involved in converting circulating effector T cells into T_{RM} cells by enhancing the expressions of key surface receptors and transcription factors required for tissue residency.[17,21] Moreover, IL-33 can promote T_{RM}-cell survival and activation through the ST2 receptor.[22] Hypoxia might also be linked to T_{RM}-cell generation and maintenance.[15] Although several mechanisms reportedly support T_{RM}-cell maintenance, little is known about the longevity of human T_{RM} cells. Notably, studies in mice and rhesus macaques demonstrate that T_{RM} cells are stable for 300 to 700 days,[23,24] suggesting that they are capable of long-term stable survival.

3. Functional characteristics of T_{RM} cells

T_{RM} cells are located in various tissues—including the skin, lungs, salivary glands, intestines, and other mucosal sites—and can exist in both lymphoid and non-lymphoid

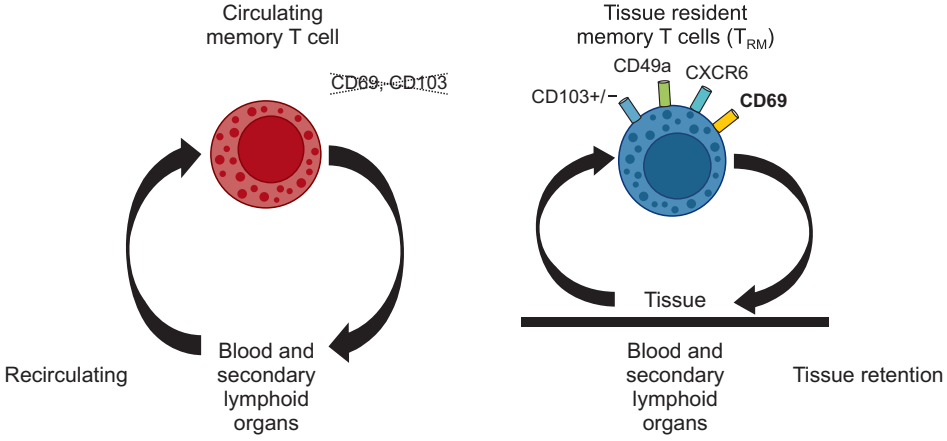

Fig. 1. Schematic representation of circulating memory T cells and tissue-resident memory T (T_{RM}) cells. Circulating memory T cells lack CD69 and CD103; recirculate between the blood and secondary lymphoid organs. T_{RM} cells express CD69 and variably express CD103, CD49a, and CXCR6, enabling their retention in tissues, which is regulated by the downregulation of KLF2 and S1PR1. CXCR6, C-X-C chemokine receptor type 6; KLF2, Krüppel-like factor 2; S1PR1, sphingosine-1-phosphate receptor 1.

tissues.[25] They are essential for conferring local immune protection against infections, and rapidly respond to pathogens without requiring recruitment from the bloodstream, thereby serving as an immediate defense line.[25] Upon encountering pathogens, T_{RM} cells can proliferate and secrete effector cytokines, such as interferon-γ (IFN-γ) and tumor necrosis factor (TNF), which control infections and recruit other immune cells to the infection site.[26,27] T_{RM} cells also contain cytotoxic molecules that can directly kill target cells.[28] In particular, T_{RM} cells play a significant role in defense against viral infections. For instance, influenza-specific T_{RM} cells in the lungs provide long-term protection by swiftly responding to re-infection and mounting a robust immune response.[29] T_{RM} cells also participate in defenses against bacterial and fungal infections. They can recognize and respond to these pathogens, ensuring rapid clearance and preventing widespread infection.[30] Importantly, T_{RM} cells adapt to the specific tissue environments they inhabit. For instance, lung T_{RM} cells are particularly adept at responding to respiratory pathogens, while skin T_{RM} cells are suited to handling cutaneous infections.[31,32] T_{RM} cells also significantly contribute to tumor surveillance and control. Higher T_{RM}-cell frequencies in tumors correlate with better patient outcomes, because these cells can produce effector cytokines and directly lyse tumor cells in various cancer types.[33] T_{RM} cells might also play a fundamental role in surveilling subclinical tumors and thereby maintaining cancer–immune equilibrium. A previous study in a mouse model demonstrated that T_{RM} cells within the epidermal layer of skin promoted a durable melanoma–immune equilibrium.[34] Skin T_{RM} cells played a crucial role in melanoma suppression, as evidenced by the findings that T_{RM}-cell generation was correlated with macroscopic tumor-free status, while T_{RM}-cell depletion led to tumor growth.

Although protective against infections, T_{RM} cells can also contribute to the pathology of autoimmune diseases. In conditions like psoriasis and vitiligo, T_{RM} cells drive inflammation and tissue damage through pro-inflammatory cytokine secretion.[27,35-37] The persistence of T_{RM} cells in affected tissues can promote chronic inflammation, exacerbating autoimmune conditions. Understanding these dual roles will help us to develop therapies that target pathogenic T_{RM} cells while preserving their protective functions. Notably, after organ transplantation, donor-derived T_{RM} cells can persist in grafts, and may either promote graft acceptance or contribute to rejection, depending on interactions between donor and recipient immune cells.[38,39] These findings suggest that T_{RM} cells might play dual roles in tissue protection and damage, which may vary depending on the clinical situation. In the following sections, we will review unique characteristics of liver T_{RM} cells, and their clinical relevance in various liver diseases.

CHARACTERISTICS OF LIVER T_{RM} CELLS

Fig. 2 presents the compositions of the mononuclear cell populations of the liver and peripheral blood, as we demonstrated in a study using liver perfusate.[40] Importantly, the liver exhibits higher proportions of natural killer cells, mucosal-associated invariant T cells, and $CD8^+$ T cells, whereas the peripheral blood shows a higher proportion of $CD4^+$ T cells, suggesting that the liver constitutes a unique immune environment. $CD69^+CD8^+$ liver T_{RM} cells comprise 20% to 80% of liver $CD8^+$ T cells.[40] Below we will summarize the unique characteristics of liver T_{RM} cells.

1. Location of liver T_{RM} cells

In most organs, T_{RM} cells reside within epithelial tissues or parenchyma; however, liver T_{RM} cells are mainly located within the sinusoids and constantly patrol the hepatic vasculature. The liver receives blood from both arterial and venous circulations, and the portal vein transports a

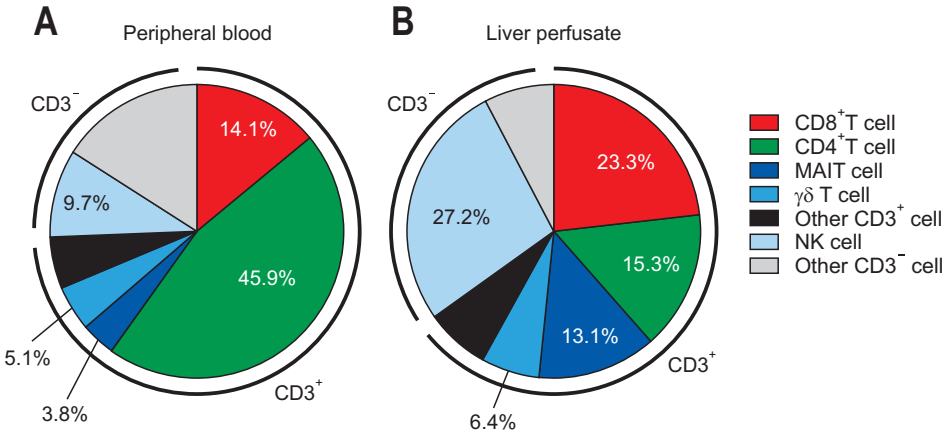

Fig. 2. Comparison of the mononuclear cell populations in peripheral blood and liver sinusoidal perfusates. Pie charts indicate the proportions of different immune cell subsets in peripheral blood (A) and liver sinusoidal perfusate (B), as measured by flow cytometry. MAIT, mucosal-associated invariant T cells; NK, natural killer cells.

significant volume of blood to the liver from the gastrointestinal tract and spleen. Upon reaching the liver, blood travels through narrow vascular capillaries called liver sinusoids, which slow the flow rate, enabling resident cells to interact with a wide range of antigens and circulating cells.[4] The liver sinusoids are lined with a thin fenestrated layer of LSECs that separates hepatocytes from circulating cells. The fenestrae allow T cells in the blood to directly access the surface of hepatocytes or tissue stroma, facilitating antigen recognition and effector functions.[41,42] In summary, liver T_{RM} cells are located within the sinusoids and are continuously exposed to the bloodstream, which may affect their unique phenotypes and functions, compared to T_{RM} cells in other tissues that are anatomically isolated from the circulation.[43] Interestingly, intravital imaging has directly shown that liver T_{RM} cells have an amoeboid shape and are uniquely positioned in the vasculature, where they patrol the liver sinusoids at faster migration speeds compared to skin T_{RM} cells.[43-45]

2. Formation of liver T_{RM} cells

During an immune response, local antigen presentation and inflammation significantly impact T_{RM}-cell differentiation and seeding within tissues.[43,46] However, the liver's vasculature ensures that circulating T_{RM}-cell precursors have direct access to liver T_{RM}-cell niches without having to leave the bloodstream. Therefore, T_{RM} cells are formed by local $CD8^+$ T-cell proliferation, but can also be induced by adhesion molecules or chemokines. The retention of circulating T cells within liver sinusoids is initially facilitated by their docking to platelets, which adhere to sinusoidal hyaluronan in a CD44-dependent manner. Next, these T cells migrate along the liver sinusoids and recognize hepatocellular antigens, which can induce liver T_{RM}-cell differentiation.[41] Another study reported that T cells may become trapped within liver sinusoids by LSECs, Kupffer cells, and hepatic stellate cells (HSCs), which promote increased expression of adhesion molecules, such as ICAM-1, VCAM-1, and VAP-1.[47] As T cells become trapped and migrate within the sinusoids, they interact with other cell types in the liver, which provide T_{RM}-inducing factors. Interactions with integrins, as well as chemokine receptors and their ligands, are crucial for liver T_{RM} cells. For example, CXCR6-CXCL16 interaction is essential for liver T_{RM}-cell retention, as is the interaction between LFA-1 and ICAM-1.[43,46,48]

3. A unique functional characteristic of liver T_{RM} cells

The liver is well-known as an immune-tolerant organ, and this concept can also be applied to liver T-cell responses. Before the concept of T_{RM} cells emerged, investigations focused on liver T-cell characteristics, particularly their trapping, activation, and tolerance mechanisms. One early study provides a concise overview of liver T-cell responses, noting that activated T cells were trapped in the liver and subsequently underwent apoptosis, indicating that the liver not only accumulates T cells but can also promote their tolerance.[49]

Under normal conditions, various gut-derived substances enter the liver through the portal vein, and the hepatic microenvironment influences liver T cells to become tolerant. To limit liver T-cell responses, HSCs express programmed death-ligand 1 (PD-L1), which triggers T-cell apoptosis.[50] Furthermore, antigen presentation by LSECs can induce antigen-specific T-cell tolerance via PD-1/PD-L1 interaction.[51] Mouse HSCs can disrupt $CD8^+$ T cells in an ICAM-1-dependent manner, thereby preventing their activation by antigen-presenting cells, and leading to apoptosis.[47] Hepatocytes can also prime $CD8^+$ T cells, but they induce clonal T-cell deletion through a Bcl-2-interacting mediator of cell death-dependent pathway.[52]

A recent study clearly demonstrated the role of LSECs in restricting liver T_{RM}-cell activation and function in a pre-clinical model of hepatitis B virus (HBV).[53] HBV-specific liver T_{RM} cells exhibited reduced function, which was induced by the adenylyl cyclase–cAMP–PKA axis and related to close contact with LSECs, suggesting that LSECs play a direct role in T-cell tolerance. Overall, these interactions between liver T cells and other cell populations contribute to the regulation of tolerance, which may differ among various clinical situations.

4. Comparisons between human and murine liver T_{RM} cells

Human and murine liver T_{RM} cells share several core characteristics. Firstly, they both exhibit upregulation of CD69, a representative marker of T_{RM} cells. This might reflect common mechanisms for retaining these cells within the liver sinusoids, and facilitating their interaction with antigens.[43,54] CXCR6 is also highly expressed in liver T_{RM} cells from both species. This chemokine receptor plays critical roles in the adhesion, accumulation, and maintenance of intrahepatic T cells. CXCL16, the ligand for CXCR6, is expressed by LSECs, Kupffer cells, and hepatocytes, which facilitates the residency of liver T_{RM} cells.[40,55,56] Moreover, in both humans and mice, liver T_{RM} cells are essential for mounting protective immune responses by producing cytokines (e.g., IFN-γ and TNF) and expressing cytotoxic molecules (e.g., granzyme B), although the expression levels may vary.[40,43] Additionally, both human and murine liver T_{RM} cells are critically influenced by IL-15, which is crucial for their development, maintenance, and homeostatic proliferation.[40,56,57]

However, there are notable differences between human and murine liver T_{RM} cells, reflecting species-specific adaptations and functions. One striking difference is that murine liver T_{RM} cells do not express CD103, which is typically found in T_{RM} cells from other tissues, and is expressed by a subset of human liver T_{RM} cells (approximately 12.4%), indicating a species-specific divergence in the phenotypic characteristics of liver T_{RM} cells.[40,43,45,56] Among human liver T_{RM} cells, $CD103^+$ cells produce more IFN-γ and IL-2 upon stimulation and express higher perforin levels, while $CD103^-$ cells (although more numerous) show less cytokine production per cell and higher PD-1 expression.[40] The expression of hypoxia-inducible factor-2α in human $CD103^-$ T_{RM} cells suggests unique regulatory mechanisms driven by the liver's hypoxic environment, which has not been prominently reported in murine studies.[40,58] These differences highlight the importance of human-specific studies to fully understand human liver T_{RM} cells, and their implications for liver diseases and therapies.

LIVER T_{RM} CELLS IN HUMAN LIVER DISEASES

Previous studies demonstrate that liver T_{RM} cells have special characteristics within the liver immune environment. In particular, their tolerant nature might play different roles in various liver diseases. Table 1 presents direct evidence regarding the liver T_{RM} population and its clinical relevance in human liver diseases. In the following sections, we will summarize the clinical and experimental studies of this topic, which reveal the dual roles of liver T_{RM} cells.

1. Viral hepatitis

T-cell responses are critical determinants of the clinical outcome of chronic HBV infection, and studies have also investigated liver T_{RM} cells' roles and relationship with clinical outcomes. Murine models have provided clues indicating that liver T_{RM} cells are a potential target for treating chronic HBV infection. Interestingly, hepatic priming of intrahepatic $CD8^+$ T cells induces dysfunctional responses, which can be restored by IL-2 treatment but not by anti-PD-L1 blockade.[59] Another study highlighted that the CXCL13-mediated accumulation of intrahepatic $CXCR5^+CD8^+$ T cells was correlated with decreased HBsAg levels, suggesting that liver T_{RM} cells may play a positive role in controlling HBV infection.[60]

Studies have also characterized the protective role of human liver HBV-specific $CD8^+$ T_{RM} cells in patients with chronic HBV infection.[56] Over 80% of liver HBV-specific $CD8^+$ T cells express CD69, and the $CD69^+CD103^+CD8^+$ subpopulation inversely correlates with HBV viral load, indicating a potential role in HBV control. This subpopulation also produces high IL-2 levels upon HBV-peptide stimulation, which may enhance HBV-specific T-cell responses. On the other hand, compared to $CD103^+$ cells, our previous study demonstrated that $CD69^+CD103^-CD8^+$ T_{RM} cells produce lower cytokine levels per cell upon HBV-peptide stimulation, although we did not investigate any direct correlations between clinical parameters.[40] Since the majority of human liver T_{RM} cells exhibit the $CD103^-$ phenotype, understanding the hypofunction mechanisms and enhancing the function of $CD69^+CD103^-CD8^+$ T_{RM} cells could be pivotal for HBV control.

The pathological roles of liver T_{RM} cells during HBV infection also warrant attention. A recent study identified highly activated liver $CD8^+$ T_{RM} cells that were associated with liver damage in chronic hepatitis B patients.[61] Upon *in vitro* stimulation with IL-2 and IL-12, these cells lysed target cells via FAS-FASL engagement, suggesting that bystander activation of T_{RM} cells during chronic HBV infection could be associated with eventual development of liver fibrosis and cirrhosis. This finding is reminiscent of the

Table 1. Human Studies of Liver T_{RM} Cells and their Clinical Correlations in Various Liver Diseases

Author	Diseases	T_{RM} phenotype	Clinical correlations from human subjects	Protective/pathologic
Pallett *et al.*[56]	HBV	$CD69^+CD103^+CD8^+$	T_{RM}↑ → HBV viral load ↓	Protective
Koda *et al.*[66]	MASLD	$CD69^+CD8^+$	T_{RM}↑ → Fibrosis↓	Protective
Nkongolo *et al.*[61]	HBV	$CXCR6^+CD8^+$	Resolution of hepatitis → T_{RM}↓	Pathologic
Kefalakes *et al.*[63]	HDV	$CD69^+CXCR6^+CD8^+$	NKG2D↑ on T_{RM} → Liver enzymes and APRI↑	Pathologic
Dudek *et al.*[65]	MASLD	$CXCR6^+CD8^+$	T_{RM}↑ → ALT↑	Pathologic
You *et al.*[67]	AIH	$CD69^+CD103^+CD8^+$	T_{RM}↑ → ALT, histologic inflammation/fibrosis↑	Pathologic
Huang *et al.*[70]	PBC	$CD103^+CD8^+$	T_{RM}↑ → ALP, GGT, TB↑, histologic inflammation/fibrosis↑	Pathologic
Kim *et al.*[40]	LC	$CD69^+CD103^-CD8^+$	T_{RM} activation↑ → MELD, Child-Pugh score↑	Pathologic

T_{RM}, tissue-resident memory T cells; HBV, hepatitis B virus; MASLD, metabolic-associated steatotic liver disease; HDV, hepatitis D virus; NKG2D, natural killer group 2D; APRI, aspartate transaminase (AST)-to-platelet ratio index; ALT, alanine aminotransferase; AIH, autoimmune hepatitis; PBC, primary biliary cholangitis; ALP, alkaline phosphatase; GGT, gamma-glutamyl transferase; TB, total bilirubin; LC, liver cirrhosis; MELD, Model for End-Stage Liver Disease.

bystander activation of $CD8^+$ T cells, which is associated with liver damage in acute hepatitis A virus infection.[62] We demonstrated that IL-15 activated non-hepatotropic virus-specific liver T_{RM} cells,[40] suggesting that the bystander activation of liver T_{RM} cells contributes to liver damage in viral hepatitis. This phenomenon has also been observed in chronic hepatitis D virus infection.[63]

2. Metabolic-associated steatotic liver disease

As observed in chronic HBV infection, liver T_{RM} cells also play dual roles in metabolic-associated steatotic liver disease (MASLD)—both promoting fibrotic processes and aiding in fibrosis resolution. Activated T_{RM} cells produce multiple cytokines, which are notably elevated in the liver and visceral fat of obese patients, potentially contributing to the inflammatory environment of the liver, and suggesting a possible association between liver damage and liver T_{RM} cells in MASLD patients.[64] This hypothesis has been elegantly proven by a recent study of human samples and mice.[65] It was demonstrated that MASLD patients exhibited elevated numbers of $CD103^+$ or $CXCR6^+$ T_{RM} cells, which displayed high surface levels of PD-1, but retained strong effector functions, with IL-15-induced production of IFN-γ, TNF, and granzyme B. These liver T_{RM} cells contributed to liver damage through non-specific cytotoxicity towards hepatocytes, especially upon downregulation of the transcription factor FOXO1. Overall, these findings indicate that the self-destructive behavior of $CD8^+$ T cells is governed by mechanisms different from those involved in antigen-specific killing by $CD8^+$ T cells.

On the other hand, a recent study found that $CD69^+CD103^-CD8^+$ T_{RM} cells may play a protective role in resolving liver fibrosis in MASLD. $CD69^+CD103^-CD8^+$ T_{RM} cells could contribute to fibrosis resolution by inducing apoptosis of HSCs. Accordingly, adoptive transfer of these cells protected mice from fibrosis progression in a CCR5-dependent manner.[66] Further studies are needed to explore the dual roles of T_{RM} cells in MASLD, and their specific mechanisms. Additionally, the role of liver T_{RM} cells in alcoholic liver disease remains to be elucidated.

3. Autoimmune liver disease

Liver T_{RM} cells also reportedly play a pathologic role in autoimmune liver diseases. In patients with autoimmune hepatitis, liver tissue exhibits high absolute numbers of $CD8^+$ T_{RM} cells, which correlate with inflammation severity and fibrosis stage.[67] Additionally, IL-15 and TGF-β appear to support liver T_{RM}-cell maintenance and survival, as their intrahepatic expression is correlated with the number of these cells. In autoimmune hepatitis patients, glucocorticoid treatment reduces hepatic inflammation, and leads to decreased numbers of liver T_{RM} cells in tissue samples. Moreover, *in vitro* glucocorticoid treatment inhibits the expansion of T_{RM} cells induced by IL-15 and TGF-β, and leads to downregulated transcriptional activity of the *BLIMP-1* gene.

Interestingly, a recent study of the biliary immune atlas revealed the presence of $CD8^+$ T_{RM} cells in regions of biliary inflammation among patients with primary sclerosing cholangitis.[68] Another study demonstrated the expansion of liver $CD4^+$ T_{RM} cells expressing genes associated with tissue residency, which are predisposed to polarize to Th17 cells.[69] In patients with primary biliary cholangitis, the frequency of $CD8^+$ T_{RM} cells is positively correlated with cholestatic liver enzymes, histologic severity (in terms of inflammation and fibrosis), and responses to ursodeoxycholic acid.[70]

4. Other liver diseases

In the context of liver cirrhosis, our investigations suggest that activation of $CD69^+CD103^-$ T_{RM} cells correlates with impaired liver function.[40] Similar to lung T_{RM} cells inducing chronic lung fibrosis after viral pneumonia,[71] liver T_{RM} cells might be involved in the development of liver fibrosis or cirrhosis in chronic HBV infection.

In organ transplant recipients, small numbers of donor cells can reportedly persist in allografts for over a decade, including $CXCR3^{hi}$ $CD8^+$ T_{RM} cells in liver transplants.[72] These cells were also present in local lymph nodes, but did not egress into the hepatic vein. The presence of long-lived T_{RM}-cell populations in liver allografts may have implications regarding liver transplantation; however, their role in rejection or other pathologic states remains to be elucidated.

5. Liver stage of malaria

Liver $CD8^+$ T_{RM} cells are pivotal in the immune defense against the liver stage of malaria. Murine studies of liver-stage malaria have provided mechanistic insights regarding the generation and maintenance of liver $CD8^+$ T_{RM} cells, which provide immediate protection by patrolling the liver sinusoids and quickly responding to sporozoite infections. Immunization with radiation-attenuated sporozoites activates liver $CD8^+$ T_{RM} cells, which are crucial for sterile immunity against malaria.[43] Moreover, depletion of these cells results in loss of protective immunity, emphasizing their importance.[43] Studies of non-human primates show that intravenous immunization with attenuated sporozoites induces parasite-specific $CD8^+$ T_{RM} cells in the liver, conferring protection similar to that observed in murine models.[73] These findings suggest that the mechanisms identified in animal studies can guide the development of

effective malaria vaccines. Future research should focus on optimizing vaccination strategies to enhance $CD8^+$ T_{RM}-cell generation and function in humans, which may lead to development of highly effective malaria vaccines.

6. Study of liver T_{RM} cells using clinical specimens

Immune cells are isolated from liver tissue samples—such as percutaneous core-needle biopsy or surgical specimens—using a combination of enzymatic and mechanical dissociation.[74,75] Briefly, fresh liver tissues are treated with enzymes, e.g., collagenase and DNase, to break down the extracellular matrix. The resulting suspension is gently homogenized, and then separated by density gradient centrifugation. After centrifugation, the immune cell layer is carefully collected, washed, and resuspended in culture medium. This immune cell population includes liver T_{RM} cells, and can be used for analyses of liver T_{RM} cells, as previously described.[40,56]

Fine-needle aspiration (FNA) offers several advantages over traditional needle biopsy, including that FNA is less invasive, better tolerated, and allows for repeated longitudinal sampling. During the procedure, a thin 22-gauge spinal needle is inserted into the liver parenchyma, and cells are aspirated with gentle negative pressure. FNA sample preparation typically involves collecting the aspirate in a culture medium, and centrifuging it to obtain a cell pellet, followed by treatment with red blood cell lysis buffer before analysis.[76] Importantly, it has been demonstrated that FNA reliably samples liver T_{RM} cells, although at slightly lower frequencies compared to in core-needle biopsy samples.[76]

Liver perfusate collection offers an alternative method for isolating intrahepatic immune cells, including liver T_{RM} cells.[77-79] During liver transplantation, graft livers are perfused with a preservation solution. The perfusate is collected, filtered, and centrifuged to isolate immune cells. Finally, liver sinusoidal mononuclear cells are separated using density gradient centrifugation. The main limitation of this technique is its dependence on liver transplantation procedures. Notably, it provides the significant advantage of yielding a large number of cells, which enables extensive and comprehensive analyses.

CONCLUSIONS

In this comprehensive review, we highlight the crucial roles and unique characteristics of T_{RM} cells in the liver, which exhibit distinct profiles that enable their tissue residency and functionality. Liver T_{RM} cells are uniquely situated within the liver sinusoids, enabling continuous interaction with circulating antigens for immune surveillance. In particular, we have summarized the protective and pathological roles of liver T_{RM} cells in various human liver diseases—including viral hepatitis, steatotic liver disease, and autoimmune liver disease.

Future research should focus on elucidating the detailed mechanisms governing the formation, maintenance, and function of T_{RM} cells in the liver. This work will improve our understanding of the balance between these cells' protective roles and potential contributions to pathology. Such insights could inform the development of targeted therapies aimed at enhancing the beneficial functions of T_{RM} cells, while mitigating their detrimental effects. Moreover, the potential use of liver T_{RM} cells in vaccine development, particularly for diseases like malaria, presents an exciting avenue for translational research.

In conclusion, liver T_{RM} cells constitute a critical component of hepatic immunity, and have significant implications regarding a wide range of liver diseases. Advancing our understanding of these cells will enhance our knowledge of liver immunology, as well as pave the way for novel therapeutic strategies in liver disease management.

CONFLICTS OF INTEREST

No potential conflict of interest relevant to this article was reported.

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AUTHOR CONTRIBUTIONS

J.W.H. and E.C.S. wrote, reviewed, and submitted the manuscript.

ORCID

Ji Won Han https://orcid.org/0000-0003-1456-1450
Eui-Cheol Shin https://orcid.org/0000-0002-6308-9503

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