IntechOpen

Biology of T Cells in Health and Disease

Edited by Hilal Arnouk

Biology of T Cells in Health and Disease

Edited by Hilal Arnouk

Published in London, United Kingdom

Biology of T Cells in Health and Disease
http://dx.doi.org/10.5772/intechopen.1002111
Edited by Hilal Arnouk

Contributors
Anand Kumar, Ankit Sriwastava, Dharmendra Kumar, Hilal Arnouk, Huiqin Hao, Jackson Sweeney, Ling Chen, Maria Daniela Holthausen Perico, Renata Kalfeltz, Shiya Wei, Shubha Gupta, Smriti Marwaha, Sundaram Gupta, Yanfang Gao, Yang Liu, Zhu Shen

First published in London, United Kingdom, 2025 by IntechOpen
IntechOpen is the global imprint of INTECHOPEN LIMITED, registered in England and Wales, registration number: 11086078, 167-169 Great Portland Street, London, W1W 5PF, United Kingdom

For EU product safety concerns: IN TECH d.o.o., Prolaz Marije Krucifikse Kozulić 3, 51000 Rijeka, Croatia, info@intechopen.com or visit our website at intechopen.com.

British Library Cataloguing-in-Publication Data
A catalogue record for this book is available from the British Library

Biology of T Cells in Health and Disease
Edited by Hilal Arnouk
p. cm.
Print ISBN 978-0-85466-863-2
Online ISBN 978-0-85466-862-5
eBook (PDF) ISBN 978-0-85466-864-9

Meet the editor

Hilal Arnouk, M.D., Ph.D., is an Associate Professor at the Department of Pathology at Midwestern University. Dr. Arnouk has received his education and post-doctorate training at Roswell Park Cancer Institute, the State University of New York at Buffalo, the Medical College of Georgia, and the University of Alabama at Birmingham. He has directed research studies in academia and biotech industry settings. His primary areas of expertise include Cancer Immunotherapy, Biomarker Discovery, and Precision Medicine. Additionally, Dr. Arnouk tremendously enjoys being an educator and mentor for medical and biomedical sciences students.

Contents

Preface

"Opponents confront us continually, but there is no opponent there. Enter deeply into an attack and neutralize it as you draw that misdirected force into your own sphere".
- Morihei Ueshiba

T cells are a heterogeneous group of effector immune cells that provide protection from infectious agents, such as bacteria and viruses, and contribute significantly to human health and disease. T Cells have been the subject of intense scientific research in the last few decades to understand their development, phenotypes, and functions fully.

This book presents an up-to-date overview of crucial topics in T cell biology, including T-cell development, T cell receptor diversity, central tolerance, peripheral tolerance, autoimmunity, the subsets of T cells, such as cytotoxic T lymphocytes, helper T lymphocytes, γδ T cells, regulatory T cells, the mechanisms of co-stimulation and co-inhibition of T cells, memory T cells, and the role of T cells in organ transplantation, autoimmune diseases, infectious Diseases, and vaccines.

Additionally, this collection will highlight the utility of T-cells in novel cancer immunotherapy approaches, such as cancer vaccines, chimeric antigen receptor T cell therapy, and adoptive cell therapies. These valuable insights will help advance our understanding of the immune system, the important contributions of T cells to our immune homeostasis and overall health, and the therapeutic applications of T cells in fighting autoimmunity, cancer, and infectious diseases.

I want to thank everyone at IntechOpen who helped with this publication. Finally, I dedicate this book to my family, colleagues, mentors, and students throughout my career.

Hilal Arnouk, M.D., Ph.D.,
Associate Professor,
Department of Pathology,
Chicago College of Osteopathic Medicine,
College of Dental Medicine,
Illinois, United States

Chicago College of Optometry,
Precision Medicine Program,
College of Graduate Studies,
Midwestern University,
Illinois, United States

Chapter 1

Gamma Delta T Cell: A Unique Member of the T Cell Family with a Significant Role in Cancer Immunotherapy

Jackson Sweeney, Smriti Marwaha and Hilal Arnouk

Abstract

Harnessing the potential of the immune system to treat cancers has been the goal of many scientific investigations and recent advancements in tumor immunology have allowed for cancer immunotherapy to become a reality. T lymphocytes that express the γδ TCR (γδ T cells) do not require antigen presentation by target cells. Instead, they recognize phospho-antigens that accumulate in tumors with increased activity of the mevalonate metabolic pathway. Additionally, the Natural Killer Group 2D (NKG2D) on γδ T cells recognizes stress-induced self-antigens widely expressed on cancer cells, such as the MHC Class I-like stress-associated molecules MIC-A and MIC-B or the UL-16 binding proteins ULBP-1, 2, and 3. This recognition can mediate direct cytotoxicity against tumor cells without prior antigen exposure or priming. Moreover, γδ T cells can be expanded when stimulated with IL-2 and Zoledronate. Collectively, these biological qualities of γδ T cells make them a promising option for cancer immunotherapy.

Keywords: Gamma Delta T cell, cancer immunotherapy, immuno-oncology, adoptive cell therapy, CAR-T cell therapy

1. Introduction

Cancer has posed a formidable challenge to researchers for decades. Since the earliest days of cancer research, scientists have been striving to develop treatments that specifically target cancer cells while sparing the surrounding healthy tissues. Recent advancements in cancer biology and emerging technologies suggest that this goal is on the cusp of becoming a reality. At the forefront of these advancements is immunotherapy, which harnesses the body's immune system to seek out and destroy cancer cells, leaving healthy tissues unharmed.

The underlying principle of immunotherapy is to leverage the body's natural defense mechanisms to treat cancer more effectively. However, like many other

treatments, immunotherapy has been met with variable success across different cancer types. When exposed to treatment pressures, many cancers evolve mechanisms to evade immune detection. Traditional immunotherapies often rely on Antigen-Presenting Cells (APCs) to prime αβ T lymphocytes to attack cancer cells specifically. This process involves recognizing Major Histocompatibility Complex (MHC) Class I molecules on the surface of cancerous cells. However, many cancers can downregulate the expression of MHC Class I molecules, evading αβ T lymphocytes and preventing these immune cells from exerting their cytotoxic effects [1]. Additionally, cancer cells can escape immune surveillance through various strategies, such as secreting the immunosuppressive cytokines Interleukin-10 (IL-10) and Transforming Growth Factor β (TGF-β) into the tumor microenvironment, dampening the immune response and leading to unchecked cancer growth [2–4]. This complex interplay between cancer cells and the immune system poses significant challenges to conventional immunotherapy strategies.

2. The biology of Gamma Delta T cell

To overcome challenges faced by the traditional cancer immunotherapy approaches, researchers have been exploring other creative solutions, including a subset of immune cells known as γδ T lymphocytes. These cells offer a promising approach to cancer immunotherapy as they do not rely on the traditional MHC Class I pathway for antigen recognition. γδ T lymphocytes are a unique subset of T cells characterized by their distinct γδ T-cell receptors (TCRs), which distinguish them from the more common αβ T cells. Instead of requiring antigen presentation from MHC Class I molecules, they are activated by conserved stress-induced self-antigens on cancer cells. The C-type lectin Natural Killer Group 2D (NKG2D) receptor on γδ T-cell recognizes these stress-induced ligands, such as MHC Class I-chain (MIC) molecules, MIC-A and MIC-B, along with UL-16 binding proteins ULBP-1, ULBP-2, and ULBP-3 on cancer cells directly and without the need for antigen presentation or priming [5]. Thus, granting γδ T lymphocytes the ability to elicit an immunological response akin to that of the innate immune system. Consequently, γδ T cells can identify and react to a wider variety of cancer cells, including those that have evolved defenses against other immune cell types. Moreover, as a subset of T cells, γδ T cells can also establish memory responses, enabling them to attack cancer cells upon subsequent exposures.

Human γδ T cells are divided into two subsets: Vδ1 and Vδ2 [6, 7]. The Vδ1 phenotype is primarily found in epithelial tissues, such as the skin, gut, and respiratory tract, where they play a critical role in immune surveillance, serving as first responders to cellular stress and transformation. Vδ1 T cells possess the C-type Natural Killer Group 2D (NKG2D) receptor, which recognizes stress-induced self-antigens, including MIC-A and MIC-B. These antigens are typically overexpressed on cancer cells, marking them for attack [8–10]. Stimulation of the NKG2D receptor, on γδ T cells, upon binding to these stress ligands, on cancer cells, can mediate direct cytotoxicity and lysis of tumor cells. Additionally, this interaction activates the release of anti-tumor cytokines, such as Interferon gamma (IFN-γ) and Tumor Necrosis Factor-α (TNF-α) [11–13].

Conversely, the second phenotype, Vδ2, is predominantly found in the peripheral blood. This subset typically binds with a second receptor, Vγ9, forming the Vγ9Vδ2 phenotype that circulates throughout the body. These cells can recognize non-peptidic phospho-antigens, such as isopentenyl pyrophosphate (IPP), which is a metabolite

of the mevalonate metabolic pathway, crucial for cellular metabolism and proliferation. In normal cells, IPP concentrations are low, and these cells remain undetected by Vγ9Vδ2 cells. However, in cancer cells, the mevalonate metabolic pathway is often upregulated to meet increased energy demands, leading to IPP accumulation and its recognition as a tumor antigen [14]. This metabolic adaptation presents a distinct opportunity for Vγ9Vδ2 T cells, which can identify and eradicate tumor cells via both TCR-dependent and TCR-independent mechanisms. This unique ability makes Vγ9Vδ2 T cells an attractive option for immunotherapy, especially when tumors become resistant to other therapeutic modalities.

Despite the promising capabilities of γδ T cells, one of the current challenges in using them for immunotherapy is their relatively low abundance in the human body. Normally, Vγ9Vδ2 cells constitute about 1–5% of peripheral blood T cells, which can be even lower in cancer patients. To address this pitfall, protocols were developed to expand these cells ex vivo. One successful strategy involves using agonist molecules to stimulate their proliferation. Zoledronate, which is already an FDA-approved bisphosphonate used to prevent bone fractures [15–17] can be used to expand Vγ9Vδ2 T cells from cancer patients with hepatocellular [18], colorectal [18], prostate [19], lymphoid [20], and breast cancers [21, 22]. Zoledronate's mechanism of action is to inhibit the enzyme, farnesyl diphosphate synthase (FDPS), which is part of the mevalonate metabolic pathway leading to the accumulation of IPP in cancer cells and subsequent TCR-dependent activation of Vγ9Vδ2 T cells. Similarly, Bromohydrin pyrophosphate (BrHPP) also interferes with the mevalonate metabolic pathway (**Figure 1**) [23–25].

In cancer patients, levels of γδ T cells are often low due to exposure to rigorous radiation and chemotherapy regimens, making ex vivo expansion a necessary solution. Studies have also shown that Vγ9Vδ2 T cells can be expanded from healthy

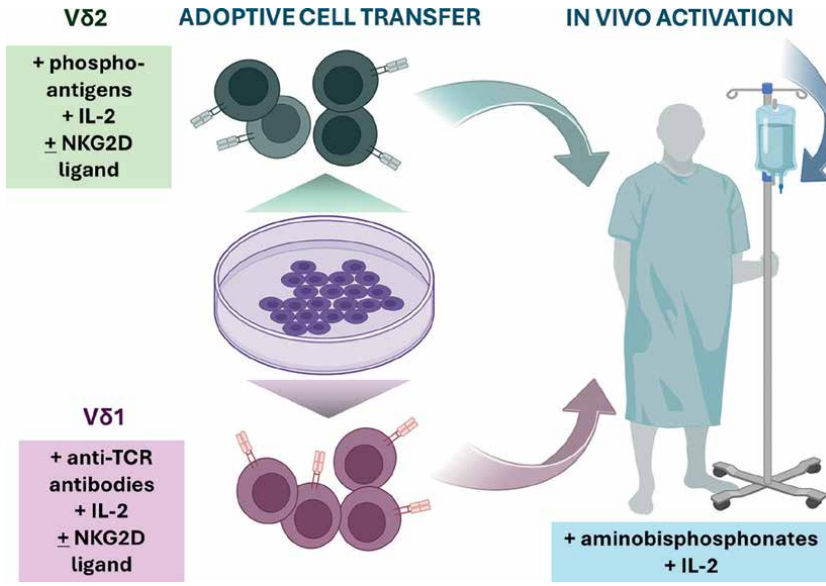

Figure 1.
Schematics showing the current strategies used in γδ T cell–based immunotherapy, including adoptive cell transfer of ex vivo-expanded γδ T cells and in vivo activation of Vδ2Vγ9 T cells by phospho-antigens. This original figure was created by the authors using Microsoft Office PowerPoint and BioRender software.

donors and transferred to cancer patients with minimal adverse effects [22, 26–28]. This is largely possible because these cells do not rely on MHC recognition for antigen detection. Consequently, donor-derived allogeneic γδ T cells are unlikely to trigger graft-versus-host disease (GVHD), a major concern in other forms of cell-based therapy.

3. Cancer immunotherapies based on γδ T cells

Research on agonist-expanded Vγ9Vδ2 T cells has been conducted across various cancer types. In colorectal cancer (CRC), Vγ9Vδ2 T cells have been identified as tumor-infiltrating lymphocytes (TILs), and their presence seems to correlate with favorable outcomes. Ex vivo expanded Vγ9Vδ2 T cells exhibit strong lytic activity against colorectal carcinoma cell lines, relying on both the TCR and the NKG2D receptor for costimulatory signaling [18, 29–31]. Interestingly, expanding Vγ9Vδ2 T cells with BrHPP or Zoledronate generates effector memory cells capable of continuously eliminating CRC cell lines upon re-exposure. Similarly, BrHPP- or Zoledronate-stimulated γδ T cells effectively lyse hepatocellular carcinoma cells while sparing healthy surrounding tissues [18]. Further research indicates that this cytotoxicity occurs through both TCR-dependent and independent mechanisms, involving interactions between the NKG2D receptor and MIC A/B, as well as between the DNAX Accessory Molecule-1 (DNAM-1) receptor and the Nectin-like molecule-5 (Necl-5) on hepatocellular carcinoma cells [32].

While expanded γδ T cells show great promise, they tend to undergo mitogen-induced apoptosis more readily than traditional αβ T lymphocytes. To address this challenge, researchers attempted expanding Vγ9Vδ2 T cells in the presence of Interleukin-12 (IL-12), which selectively protects a subset of cells from apoptosis, leading to the proliferation of these apoptosis-resistant cells. These resistant Vγ9Vδ2 T cells demonstrated effectiveness against prostate cancer cell lines, such as the DU145 and PC-3 cell lines [33, 34]. These cells appear to kill cancer cell lines via TCR-dependent interactions, as well as through interactions between Integrin Beta Chain-2 (DC18) and Intercellular Adhesion Molecule-1 (ICAM-1), facilitated by the perforin/granzyme pathway [34]. Additionally, a modified protocol for ex vivo expansion using pulsed Zoledronate stimulation was developed to mitigate the toxicity produced by inhibiting farnesyl diphosphate synthase using a continuous exposure protocol [35]. Compared to continuous exposure, the pulsed γδ T cells showed increased purity and quantities. Moreover, pulse-expanded Vγ9Vδ2 T cells produced higher levels of perforin and degranulated their granzymes in greater numbers when co-cultured with PC-3 prostate cancer cells, resulting in a 2.5-fold increase in their anti-tumor cytolytic activity [36]. In immunocompromised mice with PC-3 tumor xenotransplants, the pulsed Vγ9Vδ2 T cells reduced tumor size by 50% compared to those receiving continuously expanded Vγ9Vδ2 T cells [37].

Furthermore, the Vδ2 subset of γδ T cells has been found in mammary ductal epithelial organoids, indicating their role in immunosurveillance within these tissues. These cells produce the anti-tumor cytokine IFN-γ while efficiently killing human breast cancer cells that are triple-negative for estrogen and progesterone receptors, as well as HER2/neu [38, 39]. Additional studies have also shown that Vγ9Vδ2 T cells exhibit cytotoxic activity against various breast cancer cell lines both in vitro and in murine models. This anti-tumor activity appears to depend on breast cancer subtype, TCR engagement, and the expression of MIC A/B and ICAM-1 [40–42]. While these

results are promising, it is important to note that the predominant γδ T cell subset, Vδ1, found in TILs of breast cancer, exerts immunosuppressive effects, including the suppression of naïve T cell proliferation, dendritic cell maturation, and the secretion of immunosuppressive cytokines. The pro-tumor effects seem to be mediated by interferon gamma-induced protein 10 (IP-10), secreted by tumor cells. IP-10 recruits Vδ1 T cells to the tumor microenvironment, where they suppress anti-tumor immune responses and promote tumor growth [43].

Since ex vivo expanded Vγ9Vδ2 T cells are well-tolerated, several clinical studies have used highly enriched autologous Vγ9Vδ2 T cells prepared with Zoledronate, which were then re-infused into cancer patients to evaluate safety and potential therapeutic effects. In two early-phase clinical trials for Non-Small Cell Lung Cancer (NSCLC), stable disease was observed in three out of ten patients in one study and six out of fifteen patients in another [44, 45]. In advanced renal cell carcinoma (RCC), the transfer of γδ T cells into eleven patients resulted in prolonged tumor doubling time (DT), leading to one complete remission and stable disease in five patients [46]. Similar outcomes were achieved in metastatic RCC, where six out of ten patients showed stable disease [23]. In a clinical trial involving gastric cancer, intraperitoneal injection of ex vivo expanded Vγ9Vδ2 T cells led to a significant reduction in the volume of malignant ascites due to peritoneal dissemination in two of the seven patients enrolled [47]. In another trial, four patients with advanced refractory hematological malignancies received γδ T cells from haploidentical family donors, resulting in three complete responses [48].

Alternatively, Vγ9Vδ2 T cells can also be expanded in vivo rather than strictly ex vivo. This is typically done through the administration of the same FDA-approved amino-bisphosphonates used in ex vivo expansion, along with a low dose of IL-2. Phase I/II clinical trials demonstrated that this approach is both safe and feasible. Indeed, in vivo stimulation of Vγ9Vδ2 T cells in three breast cancer patients resulted in one case of partial remission and two cases of stable disease. This treatment was administered for over twelve months and correlated with declining levels of Cancer Antigen 15–3 (CA 15–3), a surrogate breast cancer biomarker [49]. Additionally, in vivo expansion with Zoledronate and IL-2 in nine hormone-refractory prostate cancer patients resulted in three instances of partial remission and five instances of stable disease [18]. Finally, in a trial focusing on nine patients with relapsed/refractory Non-Hodgkin's Lymphoma or multiple myeloma, partial remissions were achieved in three patients [20].

4. Conclusion

Although several clinical trials have demonstrated the safety and feasibility of γδ T cell immunotherapies, the overall response in patients has been variable and inconsistent. There might be several potential contributing factors. For instance, the agonists used in in vivo expansion may have off-target effects, such as systemic toxicity in a subset of patients. Additionally, in several clinical trials, γδ T cell infiltration and activation have been limited, possibly due to the immunosuppressive effects of the tumor microenvironment on γδ T cells [50]. There is also limited knowledge about γδ T cell checkpoint inhibition and their exact mechanisms for suppressing tumor growth. Currently, efforts are invested in developing Chimeric Antigen Receptor (CAR)-engineered γδ T cells. These genetically engineered immune cells, whether autologous or allogeneic, have the potential to be off-the-shelf cell therapy products due to their

ability to recognize antigens in an MHC-independent manner. One study successfully transduced γδ T cells with a second-generation CAR targeting GD2, demonstrating precise antitumor cytotoxicity against cancer cells expressing this specific antigen [51]. Despite the challenges and limitations observed in early clinical trials, these CAR-transduced γδ T cells could offer significant advantages as personalized cancer treatments. As the understanding of cancer biology expands and new technologies continue to develop, cancer treatments are becoming increasingly personalized, and γδ T cell therapy may be on the brink of a significant breakthrough.

Author details

Jackson Sweeney[1], Smriti Marwaha[2] and Hilal Arnouk[3]*

1 Chicago College of Osteopathic Medicine, Midwestern University, United States

2 Midwestern University Chicago College of Pharmacy, United States

3 Department of Pathology, Chicago College of Osteopathic Medicine, Precision Medicine Program, College of Graduate Studies, Chicago College of Optometry, College of Dental Medicine-Illinois, Midwestern University, United States

*Address all correspondence to: harnou@midwestern.edu

IntechOpen

References

[1] Read SB, Kulprathipanja NV, Gomez GG, Paul DB, Winston KR, Robbins JM, et al. Human alloreactive CTL inter-actions with gliomas and with those having upregulated Hla expression from exogenous IFN-gamma or IFN-gamma gene modification. Journal of Interferon & Cytokine Research. 2003;**23**(7):379-393

[2] Ksendzovsky A, Feinstein D, Zengou R, Sharp A, Polak P, Lichtor T, et al. Investigation of immunosuppressive mechanisms in a mouse glioma model. Journal of Neuro-Oncology. 2009;**93**(1):107-114

[3] Ashley DM, Kong FM, Bigner DD, Hale LP. Endogenous expression of transforming growth factor Beta1 inhibits growth and tumorigenicity and enhances Fas-mediated apoptosis in a murine high-grade glioma model. Cancer Research. 1998;**58**(2):302-309

[4] Jachimczak P, Bogdahn U, Schneider J, Beha C, Meixensberger J, Apfel R, et al. The effect of transforming growth factor-beta 2-specific phosphorothioate-anti-sense oligodeoxynucleotides in reversing cellular immunosuppression in malignant glioma. Journal of Neurosurgery. 1993;**78**(6):944-951

[5] Groh V, Rhinehart R, Secrist H, Bauer S, Grabstein KH, Spies T. Broad tumor-associated expression and recognition by tumor-derived Gamma Delta T cells of MICA and MICB. Proceedings of the National Academy of Sciences of the United States of America. 1999;**96**(12):6879-6884

[6] Hayday AC. Gamma Delta cells: A right time and a right place for a conserved third way of protection. Annual Review of Immunology. 2000;**18**:975-1026

[7] Deniger DC, Moyes JS, Cooper LJ. Clinical applications of Gamma Delta T cells with multivalent immunity. Frontiers in Immunology. 2014;**5**:636

[8] LeFranc MP, Forster A, Baer R, Stinson MA, Rabbitts TH. Diversity and rearrangement of the human T cell rearranging gamma genes: Nine germ-line variable genes belonging to two subgroups. Cell. 1986;**45**(2):237-246

[9] Forster A, Huck S, Ghanem N, Lefranc MP, Rabbitts TH. New subgroups in the human T cell rearranging V gamma gene locus. The EMBO Journal. 1987;**6**(7):1945-1950

[10] Strauss WM, Quertermous T, Seidman JG. Measuring the human T cell receptor gamma-chain locus. Science. 1987;**237**(4819):1217-1219

[11] Groh V, Steinle A, Bauer S, Spies T. Recognition of stress-induced MHC molecules by intestinal epithelial gamma delta T cells. Science. 1998;**279**(5357):1737-1740

[12] Bauer S, Groh V, Wu J, Steinle A, Phillips JH, Lanier LL, et al. Activation of NK cells and T cells by NKG2D, a receptor for stress-inducible MICA. Science. 1999;**285**(5428):727-729

[13] Saura-Esteller J, de Jong M, King LA, Ensing E, Winograd B, de Gruijl TD, et al. Gamma Delta T-cell based cancer immunotherapy: Past-present-future. Frontiers in Immunology. 2022;**13**:915837. DOI: 10.3389/fimmu.2022.915837

[14] Lafont V, Sanchez F, Laprevotte E, Michaud HA, Gros L, Eliaou JF, et al. Plasticity of γδ T cells: Impact on the anti-tumor response. Frontiers in Immunology. 2014;**5**:622

[15] Kunzmann V, Bauer E, Wilhelm M. Gamma/Delta T-cell stimulation by pamidronate. The New England Journal of Medicine. 1999;**340**(9):737-738

[16] Kondo M, Sakuta K, Noguchi A, Ariyoshi N, Sato K, Sato S, et al. Zoledronate facilitates large-scale ex vivo expansion of functional Gammadelta T cells from cancer patients for use in adoptive immunotherapy. Cytotherapy. 2008;**10**(8):842-856

[17] Sato K, Kimura S, Segawa H, Yokota A, Matsumoto S, Kuroda J, et al. Cytotoxic effects of gamma delta T cells expanded ex vivo by a third generation bisphosphonate for cancer immunotherapy. International Journal of Cancer. 2005;**116**(1):94-99

[18] Bouet-Toussaint F, Cabillic F, Toutirais O, Le Gallo M, Thomas de la Pintiere C, Daniel P, et al. Vgamma9vdelta2 T cell-mediated recognition of human solid tumors. Potential for immunotherapy of hepatocellular and colorectal carcinomas. Cancer Immunology, Immunotherapy. 2008;**57**(4):531-539

[19] Dieli F, Vermijlen D, Fulfaro F, Caccamo N, Meraviglia S, Cicero G, et al. Targeting human Gamma Delta T cells with zoledronate and Interleukin-2 for immunotherapy of hormone-refractory prostate cancer. Cancer Research. 2007;**67**(15):7450-7457

[20] Wilhelm M, Kunzmann V, Eckstein S, Reimer P, Weissinger F, Ruediger T, et al. Gamma delta T cells for immune therapy of patients with lymphoid malignancies. Blood. 2003;**102**(1):200-206

[21] Santini D, Martini F, Fratto ME, Galluzzo S, Vincenzi B, Agrati C, et al. In vivo effects of zoledronic acid on peripheral Gammadelta T lymphocytes in early breast cancer patients. Cancer Immunology, Immunotherapy. 2008;**58**(1):31-38

[22] Gaafar A, Aljurf MD, Al-Sulaiman A, Iqniebi A, Manogaran PS, Mohamed GE, et al. Defective gammadelta T-cell function and granzyme B gene polymorphism in a cohort of newly diagnosed breast cancer patients. Experimental Hematology. 2009;**37**(7):838-848

[23] Bennouna J, Bompas E, Neidhardt EM, Rolland F, Philip I, Galéa C, et al. Phase-I study of innacell gammadelta, an autologous cell-therapy product highly enriched in gamma9delta2 T lymphocytes, in combination with IL-2, in patients with metastatic renal cell carcinoma. Cancer Immunology, Immunotherapy. 2008;**57**(11):1599-1609

[24] Gober HJ, Kistowska M, Angman L, Jeno P, Mori L, De Libero G. Human T cell receptor gammadelta cells recognize endogenous mevalonate metabolites in tumor cells. The Journal of Experimental Medicine. 2003;**197**(2):163-168

[25] Kunzmann V, Bauer E, Feurle J, Weissinger F, Tony HP, Wilhelm M. Stimulation of gamma delta T cells by amino bisphosphonates and induction of anti plasma cell activity in multiple myeloma. Blood. 2000;**96**(2):384-392

[26] Bryant NL, Gillespie GY, Lopez RD, Markert JM, Cloud GA, Langford CP, et al. Preclinical evaluation of ex vivo expanded/activated γδ T cells for immunotherapy of glioblastoma multiforme. Journal of Neuro-Oncology. 2011;**101**(2):179-188

[27] Bryant NL, Suarez-Cuervo C, Gillespie GY, Markert JM, Nabors LB, Meleth S, et al. Characterization and immunotherapeutic potential of gammadelta T-cells in patients

with glioblastoma. Neuro-Oncology. 2009;**11**(4):357-367

[28] Knight A, Arnouk H, Britt W, Gillespie GY, Cloud GA, Harkins L, et al. CMV-independent lysis of glioblastoma by ex vivo expanded/activated Vδ1+ γδ T cells. PLoS One. 2013;**8**(8):e68729

[29] Corvaisier M, Moreau-Aubry A, Diez E, Bennouna J, Mosnier JF, Scotet E, et al. V Gamma 9v Delta 2 T cell response to colon carcinoma cells. Journal of Immunology. 2005;**175**(8):5481-5488

[30] Gentles AJ, Newman AM, Liu CL, Bratman SV, Feng W, Kim D, et al. The prognostic landscape of genes and infiltrating immune cells across human cancers. Nature Medicine. 2015;(8):938-945

[31] Tosolini M, Pont F, Poupot M, Vergez F, Nicolau-Travers VML, Sarry D, et al. Assessment of tumor-infiltrating TCRVγ9Vδ2 lymphocyte abundance by deconvolution of human cancers microarrays. Oncoimmunology. 2017;**6**:1-10

[32] Toutirais O, Cabillic F, Le Friec G, et al. DNAX accessory molecule-1 (CD226) promotes human hepatocellular carcinoma celllysis by γ9Vδ2 T cells. European Journal of Immunology. 2009;**39**(5):1361-1368

[33] Lopez RD, Xu S, Guo B, Negrin RS, Waller EK. CD2-mediated Il-12-dependent signals render human Gamma Delta-T cells resistant to mitogen-induced apoptosis, permitting the large-scale ex vivo expansion of functionally distinct lymphocytes: Implications for the development of adoptive immunotherapy strategies. Blood. 2000;**96**(12):3827-3837

[34] Liu Z, Guo BL, Gehrs BC, Nan L, Lopez RD. Ex vivo expanded human Vgamma9vdelta2+ gamma delta-T cells mediate innate antitumor activity against human prostate cancer cells in vitro. The Journal of Urology. 2005;**173**(5):1552-1556

[35] Wang H, Sarikonda G, Puan KJ, Tanaka Y, Feng J, Giner JL, et al. Indirect stimulation of human Vγ2vδ2 T cells through alterations in isoprenoid metabolism. Journal of Immunology. 2011;**187**(10):5099-5113

[36] Alter G, Malenfant JM, Altfeld M. Cd107a as a functional marker for the identification of natural killer cell activity. Journal of Immunological Methods. 2004;**294**(1-2):15-22

[37] Nada MH, Wang H, Workalemahu G, Tanaka Y, Morita CT. Enhancing adoptive cancer immunotherapy with Vγ2vδ2 T cells through pulse zoledronate stimulation. Journal for Immunotherapy of Cancer. 2017;**5**:9

[38] Smyth MJ, Dunn GP, Schreiber RD. Cancer immunosurveillance and immunoediting: The roles of immunity in suppressing tumor development and shaping tumor immunogenicity. Advances in Immunology. 2006;**90**:1-50

[39] Zumwalde NA, Haag JD, Sharma D, Mirrielees JA, Wilke LG, Gould MN, et al. Analysis of immune cells from human mammary ductal epithelial organoids reveals Vδ2+ T cells that efficiently target breast carcinoma cells in the presence of bisphosphonate. Cancer Prevention Research (Philadelphia, Pa.). 2016;**9**(4):305-316

[40] Bank I, Book M, Huszar M, Baram Y, Schnirer I, Brenner H. V Delta 2+ Gamma Delta T lymphocytes are cytotoxic to the MCF 7 breast carcinoma cell line and can Be detected among the T cells that infiltrate breast tumors. Clinical Immunology and Immunopathology. 1993;**67**(1):17-24

[41] Guo BL, Liu Z, Aldrich WA, Lopez RD. Innate anti-breast cancer immunity of apoptosis-resistant human gamma delta-T cells. Breast Cancer Research and Treatment. 2005;**93**(2):169-175

[42] Aggarwal R, Lu J, Kanji S, Das M, Joseph M, Lustberg B, et al. Human Vγ2vδ2 T cells limit breast cancer growth by modulating cell survival-, apoptosis-related molecules and microenvironment in tumors. International Journal of Cancer. 2013;**133**(9):2133-2144

[43] Ye J, Ma C, Wang F, Hsueh EC, Toth K, Huang Y, et al. Specific recruitment of γδ regulatory T cells in human breast cancer. Cancer Research. 2013;**73**(20):6137-6148

[44] Nakajima J, Murakawa T, Fukami T, Goto S, Kaneko T, Yoshida Y, et al. A phase I study of adoptive immunotherapy for recurrent non-small-cell lung cancer patients with autologous gammadelta T cells. European Journal of Cardiothoracic Surgery;**37**(5):1191-1197

[45] Miki S, Nakajima J, Murakawa T, Fukami T, Yoshida Y, Murayama T, et al. Adoptive immunotherapy for advanced non-small cell lung cancer using zoledronate-expanded γδ T cells: A phase I clinical study. Journal of Immunotherapy. 2011;**34**(2):202-211

[46] Kobayashi H, Tanaka Y, Yagi J, Minato N, Tanabe K. Phase I/II study of adoptive transfer of γδ T cells in combination with zoledronic acid and IL-2 to patients with advanced renal cell carcinoma. Cancer Immunology, Immunotherapy. 2011;**60**(8):1075-1084

[47] Wada I, Matsushita H, Noji S, Mori K, Yamashita H, Nomura S, et al. Intraperitoneal injection of in vitro expanded Vγ9vδ2 T cells together with zoledronate for the treatment of malignant ascites due to gastric cancer. Cancer Medicine. 2014;**3**(2):362-375

[48] Martin W, Smetak M, Schaefer-Eckart K, Kimmel B, Birkmann J, Einsele H, et al. Successful adoptive transfer and in vivo expansion of haploidentical γδ T cells. Journal of Translational Medicine. 2014;**12**(1):45

[49] Meraviglia S, Eberl M, Vermijlen D, Todaro M, Buccheri S, Cicero G, et al. In vivo manipulation of Vgamma-9vdelta2 T cells with zoledronate and low-dose Interleukin-2 for immunotherapy of advanced breast cancer patients. Clinical and Experimental Immunology. 2010;**161**(2):290-297

[50] Revesz IA, Joyce P, Ebert LM, Prestidge CA. Effective γδ T-cell clinical therapies: Current limitations and future perspectives for cancer immunotherapy. Clinical and Translational Immunology. 2024;**13**(2):e1492

[51] Capsomidis A, Benthall G, Van Acker HH, Fisher J, Kramer AM, Abeln Z, et al. Chimeric antigen receptor-engineered human Gamma Delta T cells: Enhanced cytotoxicity with retention of cross presentation. Molecular Therapy. 2018;**26**:354-365

Chapter 2

CAR-T Cells: A Breakthrough in Cancer Treatment

Ankit Sriwastava, Shubha Gupta, Anand Kumar, Sundaram Gupta and Dharmendra Kumar

Abstract

Cancer is a significant health problem that demands ongoing innovation in treatment approaches. This chapter examines the evolution of cancer therapies, highlighting the limitations of conventional methods and the need for precise and effective solutions. Chimeric Antigen Receptor T cell therapy (CAR-T) emerges as a promising solution. The chapter delves into the fundamentals of CAR-T cell therapy, explaining the process of engineering these cells and their mechanism of action. It also discusses the clinical applications of CAR-T cell therapy in approved indications for hematologic malignancies. CAR-T cell therapy has expanded its scope to solid tumors and is exploring futuristic possibilities, such as combination therapies. Although there are challenges, ongoing research focuses on enhancing accessibility. The collaborative and interdisciplinary nature of cancer treatments is emphasized.

Keywords: CAR-T, T-cells, B-ALL, DLBCL, hematologic malignancies

1. Introduction

Cancer persists as a substantial global health challenge, demanding ongoing innovation in therapeutic strategies. The contemporary framework of cancer treatment reflects a collaborative, multidisciplinary endeavor to confront the intricacies of the disease. While conventional methods like chemotherapy and radiation exhibit partial effectiveness, their utility is hampered by substantial limitations encompassing issues of both efficacy and adverse effects. As the complexities of cancer become increasingly apparent, the imperative for continual advancements in treatment approaches underscores the pressing need for more targeted and refined solutions to enhance overall efficacy and mitigate the challenges associated with traditional modalities [1]. The pressing need for targeted and effective cancer treatments has propelled the investigation of innovative approaches, with CAR-T cell therapy standing out as a promising frontier in this quest. This chapter is dedicated to unraveling the evolutionary trajectory of cancer treatment, elucidating the limitations inherent in conventional methods. By doing so, it sets the stage for a comprehensive exploration of the revolutionary CAR-T cell therapy—a cutting-edge and transformative modality that holds immense

potential in overcoming the challenges posed by traditional treatments. Through this exploration, the chapter aims to provide a nuanced understanding of the historical context and pave the way for a deeper appreciation of the advancements in CAR-T cell therapy within the broader landscape of cancer treatment [2]. The evolution of CAR-T cell therapy is intimately entwined with the historical progression of cancer therapies, particularly within the expansive domain of immunotherapy. Immunotherapy, distinguished by its emphasis on leveraging the body's own immune system to combat cancer, has traversed a transformative journey marked by pivotal milestones and groundbreaking discoveries. These key advancements have not only reshaped the landscape of oncology but have also laid the foundation for the development of novel and targeted therapies such as CAR-T cell therapy. In tracing this historical trajectory, we gain a profound appreciation for the interconnectedness of these therapeutic approaches and the profound impact they collectively wield in advancing the frontiers of cancer treatment [3]. The evolution of cancer therapies leading up to CAR-T cell therapy represents a series of scientific and clinical breakthroughs. To appreciate the significance of CAR-T cell therapy, it is essential to trace the roots of immunotherapy, acknowledging the key moments and discoveries that paved the way for this revolutionary treatment modality.

2. Understanding CAR-T cell therapy

2.1 What are CAR-T cells?

CAR-T cells, or Chimeric Antigen Receptor T cells, represent a cutting-edge form of immunotherapy designed to harness the body's immune system to combat cancer. At the heart of CAR-T therapy is the Chimeric Antigen Receptor—a synthetic receptor that combines the specificity of an antibody with the potent cytotoxicity of T cells. This receptor is engineered to recognize and bind to specific antigens expressed on the surface of cancer cells [4]. The Chimeric Antigen Receptor is typically composed of three main components: an extracellular domain, a transmembrane domain, and an intracellular signaling domain. The extracellular domain, derived from an antibody, confers the CAR-T cell with the ability to recognize a particular antigen in cancer cells. The transmembrane domain anchors the receptor in the cell membrane, while the intracellular signaling domain triggers the activation of the T cell upon antigen binding [3].

2.1.1 How CAR-T cells are engineered

CAR-T cells are engineered through a complex process that involves isolating T cells from a patient's blood, modifying them *ex vivo*, and then reinfusing them into the patient. This process begins by extracting T cells from the patient, typically through apheresis [5]. Once isolated, these T cells are genetically modified to express the Chimeric Antigen Receptor using viral vectors, often derived from lentiviruses or retroviruses [6]. The modified CAR-T cells are then expanded in culture to create a robust population. This personalized army of CAR-T cells, now armed with the synthetic receptor, is infused back into the patient. Once in the body, these engineered cells seek out and selectively destroy cancer cells expressing the targeted antigen (**Figure 1**).

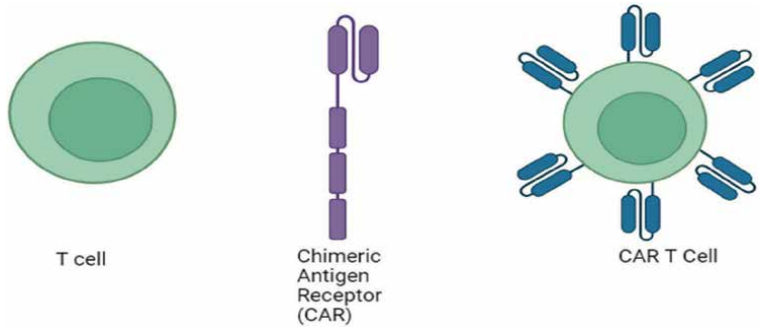

Figure 1.
CAR-T cells.

2.2 Mechanism of action

Step-by-step process of CAR-T cell therapy:

Extraction of T cells: the process begins with the extraction of T cells from the patient through apheresis, a technique that separates blood components.

Genetic modification: the isolated T cells undergo genetic modification *ex vivo*. Using viral vectors, typically derived from lentiviruses or retroviruses, the Chimeric Antigen Receptor (CAR) is introduced into the T cells. This step equips the T cells with the ability to recognize and bind to specific antigens on the surface of cancer cells.

Cell expansion: the genetically modified T cells are cultured and expanded *in vitro* to create a substantial population of CAR-T cells. This ensures a sufficient quantity of engineered cells for therapeutic effectiveness.

Infusion into the patient: the expanded CAR-T cell population is then infused back into the patient. This infusion marks the introduction of the modified T cells, now armed with the CAR, into the patient's bloodstream.

Homing to tumor sites: once in the body, CAR-T cells navigate the bloodstream and homing mechanisms to reach tumor sites. The CAR enables these T cells to specifically recognize and bind to cancer cells expressing the targeted antigen.

Activation and cytotoxicity: upon binding to cancer cells, the CAR-T cells become activated. This activation triggers the release of cytotoxic substances and the initiation of immune responses that lead to the destruction of the cancer cells.

Persistence and memory: some CAR-T cells persist in the body, forming a "memory" population. This persistence enhances the ability of the immune system to mount prolonged responses against cancer cells, offering potential long-term protection.

2.3 Interaction between CAR-T cells and cancer cells

The interaction between CAR-T cells and cancer cells is highly specific. The Chimeric Antigen Receptor on the surface of CAR-T cells acts as a molecular sensor, recognizing and binding to specific antigens present on the cancer cell surface. This interaction initiates a cascade of signaling events within the CAR-T cell, leading to its activation.

The engineered CAR-T cells unleash potent cytotoxic mechanisms, including the release of perforin and granzymes, inducing apoptosis (cell death) in the cancer cells. Additionally, the activated CAR-T cells can stimulate other components of the immune system, such as macrophages and natural killer cells, further amplifying the anti-cancer immune response (**Figure 2**) [7].

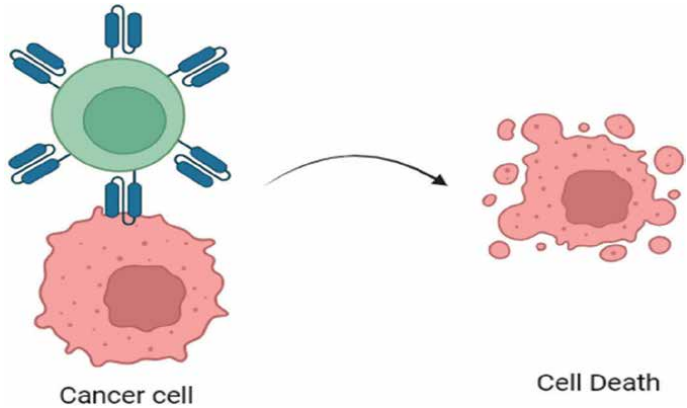

Figure 2.
Diagrammatic representation of CAR-T cell interaction with cancer cells lead to cell death.

3. Development and advancements

3.1 Early research and clinical trials

The journey of CAR-T cell therapy commenced with pioneering studies and clinical trials that laid the foundational groundwork. In the early stages, researchers focused on understanding the feasibility and safety of genetically modifying T cells to express Chimeric Antigen Receptors (CARs). Notable studies, such as those by Sadelain and Brentjens, marked crucial milestones in demonstrating the concept's viability and its potential as a transformative cancer treatment [8]. The early phases of CAR-T cell therapy research faced significant challenges, including issues related to the persistence and functionality of engineered T cells, as well as concerns about off-target effects. These challenges prompted researchers to refine the CAR design, optimize the genetic modification process, and enhance the overall therapeutic efficacy. Breakthrough moments emerged as solutions were devised, leading to improved CAR-T cell technologies and addressing initial hurdles [9].

The landmark clinical trial by Maude et al. in 2014 demonstrated unprecedented success in treating pediatric patients with acute lymphoblastic leukemia (ALL), showcasing the therapy's potential for achieving sustained remissions and transforming the landscape of cancer treatment [10].

3.2 Key developments in CAR-T cell therapy

Over the years, CAR-T cell therapy has undergone significant technological advancements, revolutionizing cancer treatment. One major breakthrough has been the refinement of CAR design. Researchers have developed next-generation CARs with enhanced signaling domains, optimized co-stimulatory molecules, and improved antigen recognition capabilities. These technological advancements have contributed to the development of CAR-T cells with increased potency and persistence in the body, leading to improved treatment outcomes [11].

Another critical development is the evolution of manufacturing processes. Streamlining and optimizing the production of CAR-T cells have become pivotal

for scalability and widespread clinical application. Innovative manufacturing technologies, such as automated systems and closed bioreactors, have been introduced to ensure consistent and high-quality production of CAR-T cell therapies [12]. Enhancing the safety profile of CAR-T cell therapy has been a key focus of research and development. Initial concerns regarding cytokine release syndrome (CRS) and neurotoxicity led to the development of strategies to mitigate these adverse events. The introduction of cytokine-blocking agents and the refinement of dosing regimens have contributed to a more favorable safety profile, making CAR-T cell therapy more manageable for patients [13].

Moreover, advancements in target antigen selection and validation have played a crucial role in improving the therapy's efficacy. Identifying antigens that are highly specific to cancer cells while sparing healthy tissues has enhanced the precision of CAR-T cell targeting, minimizing off-target effects and maximizing therapeutic efficacy [14].

4. Clinical applications

4.1 Approved indications

CAR-T cell therapy has gained approval for treating specific types of cancers, marking a standard shift in oncology. Notably, the U.S. Food and Drug Administration (FDA) has approved CAR-T therapies for certain hematologic malignancies. Among these, CD19-targeted CAR-T cells have shown remarkable success in treating B-cell acute lymphoblastic leukemia (B-ALL) and diffuse large B-cell lymphoma (DLBCL). The approval of these therapies reflects the transformative impact of CAR-T cell therapy in providing effective treatment options for patients with refractory or relapsed hematologic cancers [15, 16]. Success stories in CAR-T cell therapy are exemplified by cases of patients achieving sustained remissions and, in some instances, achieving complete responses. Notable outcomes include patients with B-ALL experiencing long-term remission following CAR-T treatment. These success stories underscore the therapeutic potential of CAR-T cell therapy, especially in cases where traditional treatments have failed [17].

4.2 Ongoing research and potential applications

Ongoing research is expanding the scope of CAR-T cell therapy beyond hematologic malignancies. Investigations are underway to explore the potential application of CAR-T cells in solid tumors, such as glioblastoma and pancreatic cancer. Researchers are adapting CAR designs and exploring new target antigens to overcome the challenges posed by the tumor microenvironment in solid tumors. This expansion into diverse cancer types represents a promising avenue for broadening the impact of CAR-T cell therapy [18, 19]. The field of CAR-T cell therapy continues to evolve with emerging trends and futuristic possibilities. These include the development of "off-the-shelf" CAR-T cells, overcoming the current limitation of personalized manufacturing for each patient. Additionally, researchers are investigating combination therapies, integrating CAR-T cells with other treatment modalities to enhance overall anti-cancer efficacy. The potential for CAR-T cells to serve as a platform for delivering therapeutic payloads or cytokines is another exciting avenue under exploration [20, 21].

5. Challenges and considerations

5.1 Side effects and toxicities

CAR-T cell therapy, while revolutionary, is associated with specific side effects and toxicities. One of the most common adverse events is Cytokine Release Syndrome (CRS), a systemic inflammatory response triggered by the rapid activation and proliferation of CAR-T cells. CRS can range from mild flu-like symptoms to severe manifestations, potentially impacting multiple organs. Neurotoxicity is another concern, characterized by cognitive disturbances and, in extreme cases, seizures. Understanding and managing these side effects are crucial for optimizing patient outcomes [13, 14]. To mitigate side effects, clinicians employ various strategies. Tocilizumab, an interleukin-6 receptor antagonist, is frequently used to counteract CRS. Corticosteroids may be administered to address severe immune reactions. Monitoring patients closely and intervening promptly are essential aspects of complication management. Ongoing research aims to develop predictive biomarkers that can identify individuals at higher risk of severe complications, enabling proactive interventions to minimize the impact of side effects [22, 23].

5.2 Cost and accessibility

CAR-T cell therapy poses economic challenges due to its intricate manufacturing process, personalized nature, and the costs associated with clinical management of side effects. Infrastructure requirements, including specialized facilities for cell processing, contribute to the overall expense. The initial costs of CAR-T therapies can be substantial, impacting healthcare systems, insurers, and patients alike. Economic implications extend beyond treatment costs to considerations of long-term healthcare expenditures associated with managing potential late effects and complications [24, 25].

Recognizing the need for broader accessibility, ongoing efforts are directed toward reducing the economic barriers to CAR-T cell therapy. This involves research into more cost-effective manufacturing methods, increased collaboration between healthcare providers and manufacturers, and negotiations to establish reimbursement models that ensure affordability for both healthcare systems and patients. Furthermore, exploring strategies for "off-the-shelf" CAR-T cells could potentially streamline production processes, enhancing accessibility by reducing the need for personalized manufacturing [26, 27].

6. Future directions and outlook

6.1 Next-generation CAR-T cell therapies

The field of CAR-T cell therapy is rapidly advancing, with ongoing research focused on developing next-generation technologies to enhance its efficacy and safety. Researchers are exploring novel CAR designs with advanced signaling domains, optimized co-stimulatory molecules, and improved antigen recognition capabilities. Additionally, advancements in gene-editing technologies, such as CRISPR-Cas9, are being harnessed to precisely engineer CAR-T cells, allowing for more controlled and

predictable modifications. Ongoing efforts aim to address current limitations, such as antigen escape and persistence of CAR-T cells, with a focus on creating more potent and durable therapies [28, 29].

The future of CAR-T cell therapy holds promise for several innovations. These include the development of "universal" or "off-the-shelf" CAR-T cells, allowing for standardized, readily available treatments without the need for personalized manufacturing. Enhanced safety features, such as suicide switches, are under investigation to provide a fail-safe mechanism for controlling CAR-T cell activity and minimizing adverse events. Moreover, advancements in delivery systems and targeting strategies are being explored to improve the precision and efficiency of CAR-T cell therapy across various cancer types [30, 31].

6.2 Integration with other therapies

Combination therapies involving CAR-T cell therapy and other treatment modalities are a burgeoning area of research. Researchers are exploring synergistic effects by combining CAR-T cells with traditional therapies like chemotherapy or radiation. Additionally, combining CAR-T cell therapy with immune checkpoint inhibitors aims to enhance the overall immune response against cancer cells. These synergistic approaches seek to capitalize on the strengths of different treatments, potentially improving response rates and extending the benefits of CAR-T cell therapy to a broader range of patients [32, 33]. Collaboration is becoming increasingly crucial in advancing cancer treatment. Interdisciplinary collaborations among immunologists, oncologists, geneticists, and other experts are fostering a comprehensive understanding of cancer biology and immune response dynamics. This collaborative approach extends beyond individual therapies, emphasizing the integration of diverse treatment modalities into cohesive and personalized cancer care strategies. Such collaborative efforts are essential for optimizing treatment outcomes and navigating the complexities of individual patient responses [34, 35].

7. Conclusion

In conclusion, CAR-T cell therapy stands as a revolutionary and transformative approach in cancer treatment. The significance of CAR-T cell therapy lies in its ability to harness the body's own immune system to target and eliminate cancer cells with precision. This therapy has demonstrated remarkable success, particularly in the treatment of certain hematologic malignancies, where conventional treatments have shown limitations. The incorporation of Chimeric Antigen Receptors into T cells represents a groundbreaking strategy that has paved the way for personalized and highly effective cancer therapies.

As of now, CAR-T cell therapy has received approvals for specific indications, showcasing its clinical success and the paradigm shift it has brought to the field of oncology. The therapy's future potential is vast, with ongoing research focusing on refining current technologies, developing next-generation CAR-T cells, and exploring innovative combination therapies. The continuous evolution of CAR-T cell therapy holds promise for extending its application to a broader spectrum of cancer types and improving its accessibility and safety.

Acknowledgements

The author acknowledges the usage of Chat GPT, Quilbolt, (AI tool used) for language polishing of the manuscript.

Author details

Ankit Sriwastava[1*], Shubha Gupta[2], Anand Kumar[1], Sundaram Gupta[3] and Dharmendra Kumar[1]

1 Department of Orthopaedic Surgery, King George's Medical University, India

2 Department of Biochemistry, AIIMS Raipur, India

3 Department of Hematology and Stem Cell Center, SGPGIMS, India

*Address all correspondence to: ankitshiv130294@gmail.com

IntechOpen

References

[1] Debela DT, Muzazu SG, Heraro KD, et al. New approaches and procedures for cancer treatment: Current perspectives. SAGE Open Medicine. 2021;**9**:20503121211034366. DOI: 10.1177/20503121211034366

[2] Mitra A, Barua A, Huang L, Ganguly S, Feng Q, He B. From bench to bedside: The history and progress of CAR T cell therapy. Frontiers in Immunology. 2023;**14**:1188049. DOI: 10.3389/fimmu.2023.1188049

[3] Gomes-Silva D, Ramos CA. Cancer immunotherapy using CAR-T cells: From the research bench to the assembly line. Biotechnology Journal. 2018;**13**(2):1-16. DOI: 10.1002/biot.201700097

[4] Pan K, Farrukh H, Chittepu VCSR, Xu H, Pan CX, Zhu Z. CAR race to cancer immunotherapy: From CAR T, CAR NK to CAR macrophage therapy. Journal of Experimental & Clinical Cancer Research. 2022;**41**(1):119. DOI: 10.1186/s13046-022-02327-z

[5] Zhang C, Liu J, Zhong JF, Zhang X. Engineering CAR-T cells. Biomarker Research. 2017;**5**:22. DOI: 10.1186/s40364-017-0102-y

[6] Warnock JN, Daigre C, Al-Rubeai M. Introduction to viral vectors. Methods in Molecular Biology. 2011;**737**:1-25. DOI: 10.1007/978-1-61779-095-9_1

[7] Maude SL, Frey N, Shaw PA, et al. Chimeric antigen receptor T cells for sustained remissions in leukemia [published correction appears in N Engl J Med. 2016 Mar 10;374(10):998]. The New England Journal of Medicine. 2014;**371**(16):1507-1517. DOI: 10.1056/NEJMoa1407222

[8] Sadelain M, Brentjens R, Rivière I. The basic principles of chimeric antigen receptor design. Cancer Discovery. 2013;**3**(4):388-398. DOI: 10.1158/2159-8290.CD-12-0548

[9] Grupp SA, Kalos M, Barrett D, et al. Chimeric antigen receptor-modified T cells for acute lymphoid leukemia. The New England Journal of Medicine. 2013;**368**(16):1509-1518. DOI: 10.1056/NEJMoa1215134

[10] Maude SL, Frey N, Shaw PA, et al. Chimeric antigen receptor T cells for sustained remissions in leukemia. The New England Journal of Medicine. 2014;**371**(16):1507-1517. DOI: 10.1056/NEJMoa1407222

[11] Park JH, Geyer MB, Brentjens RJ. CD19-targeted CAR T-cell therapeutics for hematologic malignancies: Interpreting clinical outcomes to date. Blood. 2016;**127**(26):3312-3320. DOI: 10.1182/blood-2016-03-643544

[12] Levine BL, Miskin J, Wonnacott K, Keir C. Global manufacturing of CAR T cell therapy. Molecular Therapy—Methods & Clinical Development. 2017;**15**(4):92-101. DOI: 10.1016/j.omtm.2016.12.006

[13] Neelapu SS, Tummala S, Kebriaei P, et al. Chimeric antigen receptor T-cell therapy—Assessment and management of toxicities. Nature Reviews. Clinical Oncology. 2018;**15**(1):47-62. DOI: 10.1038/nrclinonc.2017.148

[14] Teachey DT, Lacey SF, Shaw PA, et al. Identification of predictive biomarkers for cytokine release syndrome after chimeric antigen receptor T-cell therapy for acute lymphoblastic leukemia. Cancer Discovery. 2016;**6**(6):664-679. DOI: 10.1158/2159-8290.CD-16-0040.*

[15] Maude SL, Laetsch TW, Buechner J, et al. Tisagenlecleucel in children and young adults with B-cell lymphoblastic leukemia. The New England Journal of Medicine. 2018;**378**(5):439-448. DOI: 10.1056/NEJMoa1709866

[16] Schuster SJ, Bishop MR, Tam CS, et al. Tisagenlecleucel in adult relapsed or refractory diffuse large B-cell lymphoma. The New England Journal of Medicine. 2019;**380**(1):45-56. DOI: 10.1056/NEJMoa1804980

[17] Park JH, Rivière I, Gonen M, et al. Long-term follow-up of CD19 CAR therapy in acute lymphoblastic leukemia. The New England Journal of Medicine. 2018;**378**(5):449-459. DOI: 10.1056/NEJMoa1709919

[18] Brown CE, Mackall CL. CAR T cell therapy: Inroads to response and resistance. Nature Reviews. Immunology. 2019;**19**(2):73-74. DOI: 10.1038/s41577-019-0123-7

[19] Rafiq S, Hackett CS, Brentjens RJ. Engineering strategies to overcome the current roadblocks in CAR T cell therapy. Nature Reviews. Clinical Oncology. 2020;**17**(2):147-167. DOI: 10.1038/s41571-019-0278-6

[20] Fraietta JA, Lacey SF, Orlando EJ, et al. Determinants of response and resistance to CD19 chimeric antigen receptor (CAR) T cell therapy of chronic lymphocytic leukemia. Nature Medicine. 2018;**24**(5):563-571. DOI: 10.1038/s41591-018-0010-1

[21] Bonifant CL, Jackson HJ, Brentjens RJ, Curran KJ. Toxicity and management in CAR T-cell therapy. Molecular Therapy—Oncolytics. 2016;**31**(3):16011. DOI: 10.1038/mto.2016.11.

[22] Lee DW, Santomasso BD, Locke FL, et al. ASTCT consensus grading for cytokine release syndrome and neurologic toxicity associated with immune effector cells. Biology of Blood and Marrow Transplantation. 2019;**25**(4):625-638. DOI: 10.1016/j.bbmt.2018.12.758

[23] Brudno JN, Kochenderfer JN. Recent advances in CAR T-cell toxicity: Mechanisms, manifestations and management. Blood Reviews. 2019;**34**:45-55. DOI: 10.1016/j.blre.2018.10.002

[24] Makita S, Yoshimura K, Tobinai K. Economic evaluation of chimeric antigen receptor T-cell therapy. Current Treatment Options in Oncology. 2020;**21**(12):102. DOI: 10.1007/s11864-020-00784-2

[25] Pearson S, Cordon-Cardo C, Cohen D, et al. Institutional considerations in leveraging CAR T-cell therapy for patients with cancer during the COVID-19 pandemic. JAMA Oncology. 2020;**6**(8):1131-1134. DOI: 10.1001/jamaoncol.2020.3051

[26] Puthenveetil G, Musto S, Rodriguez-Molina J, et al. Cost-effectiveness and budget impact of chimeric antigen receptor T-cell therapy in relapsed or refractory large B-cell lymphoma. Value in Health. 2021;**24**(6):826-834. DOI: 10.1016/j.jval.2020.11.021

[27] Bach PB, Giralt S, Saltz L. FDA approval of Tisagenlecleucel: Promise and complexities of a $475 000 cancer drug. JAMA. 2017;**318**(23):1861-1862. DOI: 10.1001/jama.2017.17127.*

[28] Majzner RG, Mackall CL. Clinical lessons learned from the first leg of the CAR T cell journey. Nature Medicine. 2019;**25**(9):1341-1355. DOI: 10.1038/s41591-019-0502-y

[29] June CH, Sadelain M. Chimeric antigen receptor therapy. The

New England Journal of Medicine. 2018;**379**(1):64-73. DOI: 10.1056/NEJMra1706169

[30] Gill S, Maus MV, Porter DL. Chimeric antigen receptor T cell therapy: 25 years in the making. Blood Reviews. 2016;**30**(3):157-167. DOI: 10.1016/j.blre.2015.10.003

[31] Zhao Z, Condomines M, van der Stegen SJC, et al. Structural design of engineered costimulation determines tumor rejection kinetics and persistence of CAR T cells. Cancer Cell. 2015;**28**(4):415-428. DOI: 10.1016/j.ccell.2015.09.004

[32] Rafiq S, Yeku OO, Jackson HJ, et al. Targeted delivery of a PD-1-blocking scFv by CAR-T cells enhances anti-tumor efficacy in vivo. Nature Biotechnology. 2018;**36**(9):847-856. DOI: 10.1038/nbt.4195

[33] Adusumilli PS, Zauderer MG, Rusch VW, et al. Regional delivery of mesothelin-targeted CAR T cells for pleural cancers: Safety and preliminary efficacy in combination with anti-PD-1 agent. Journal of Clinical Oncology. 2019;**37**(Suppl. 15):2511. DOI: 10.1200/JCO.2019.37.15_suppl.2511

[34] Sharma P, Hu-Lieskovan S, Wargo JA, Ribas A. Primary, adaptive, and acquired resistance to cancer immunotherapy. Cell. 2017;**168**(4):707-723. DOI: 10.1016/j.cell.2017.01.017

[35] Ribas A, Wolchok JD. Cancer immunotherapy using checkpoint blockade. Science. 2018;**359**(6382):1350-1355. DOI: 10.1126/science.aar4060

Chapter 3

T-Cell Receptor: From T-Cell Function to T-Cell Clonality

Maria Daniela Holthausen Perico and Renata Kalfeltz

Abstract

Evaluation of T cell clonality has been costly and/or time-consuming. The analysis of TCR β-chain constant region 1 (TRBC1) provides a simplified immunophenotypic assessment of T-cell clonality. Furthermore, due to the high variability of T-cell populations, there is a need for reliable and robust panels to sort normally from pathological T-cells. The CD27 and CD45RA phenotypic profiling strategy associated with the evaluation of TCRCBeta1 in the same cytometry tube is able to separate normal T Cell populations from clonal populations, gating clusters of cells according to their CD45RA x CD27 expression and then evaluate their TCRCBeta1 status. TCRCBeta1 marker is not only easily implemented in routine immunophenotyping but is also faster and much cheaper than the analysis of TCR-VBeta families either by PCR or by flow cytometry.

Keywords: TCRCBeta1, T cell neoplasm diagnosis, flow cytometry, immunophenotyping, clonality

1. Introduction

Flow cytometry is a powerful tool for analyzing lymphoid subsets. This technology evolved from the simple quantification of TCD4 and TCD8 cells in HIV-positive patients to a broader comprehension of the lymphoid compartment. Recent publications describe impressive values as high as 85 different types of TCD4 and 45 types of TCD8.

The number of different T-cell phenotypes according to their functions makes the clonality assessment of T-cell populations even more challenging.

For a very long time, flow cytometry specialists all around the world waited for a so-to-speak "T cell kappa/lambda," meaning a marker that could easily identify clonal T Cell populations with accuracy.

It turns out that the β-chain of T-cell receptor αβ (TCR) structure has a variant and a constant region. The variability of the constant region? It is region 1 or region 2 as simple as that.

Recently, screening of anti-TCR monoclonal antibodies revealed a high specificity of clone JOVI-1 for the TCR β-chain constant region 1 (TCRBC1) domain, providing an opportunity for a simplified immunophenotypic assessment of T-cell clonality. There are two genes associated with the β-chain constant region: TCRBC1 and

TCRBC2. Each TCR (and therefore each T cell) irreversibly selects a TCR β-chain constant region encoded by either TCRBC1 or TCRBC2 for expression in a mutually exclusive manner, similar to the kappa and lambda immunoglobulin light chain utilization by B-cells. Therefore, normal TCRαβ T-cell populations are expected to exhibit comparable numbers of TCRBC1-positive and TCRBC2-positive subsets [1].

Since the vast majority of T-cell malignancies derive from the largely dominant TCRαβ T-cell subset, this approach is applicable to most scenarios where a neoplastic T-cell population is suspected.

2. From T-cell function to T-cell clonality

2.1 Back to basics: a brief recap about the immune system

The cellular component of the innate (or natural) immune system consists of all the cells that lack immunologic memory, have phagocytic properties or release inflammatory mediators, such as neutrophils, eosinophils, monocytes, and natural killer cells.

The acquired (or adaptive) immune response is probably one of the most complex and advanced systems in known biology, with the ability to identify and memorize virtually any foreign antigen. It involves the proliferation of antigen-specific B and T cells, as well as their complex interactions.

While the innate response occurs to the same extent regardless of how many times the antigen is encountered, the acquired response improves with repeated exposure (1).

Both systems are deeply and beautifully intertwined: activated innate immune cells convey the information about the nature and origin of the antigen to the adaptive immune cells, which will elaborate the proper and specific response [2–5].

Flow cytometry identifies the major types and subtypes of lymphocytes.

2.2 T cells

T cells are a heterogeneous group of short- and long-lived cells.

Under normal circumstances, the long-lived cells, typically contained within the *naïve* subset, are quiescent, remaining in a non-cycling state for months to years while awaiting encounter with cognate antigen [2–5]. The short-lived cells are generally contained within the effector and memory subsets, undergoing variable levels of cell cycling in response to antigens encountered throughout the lifetime of the host.

2.3 Sorting out different types of T-cells: the CD45RA and CD27 strategy

The major T Cell populations divide into *naive* T cells (those that have not yet contacted foreign antigens), effector T cells (that will disappear once the antigen is eliminated), and memory T cells (that may survive for years and may be easily reactivated if the same antigen appears again) (3). The *naive* and memory cells can be distinguished by the expression of different versions of the CD45 molecule: *naive* cells express CD45RA, and memory cells express CD45RO (1); these antigens can help identify different T cell subsets: *naive* (CD27+ CD45RA+), central/transitional memory (CM/TM CD27+ CD45RA negative), effector memory (EM CD27 negative and CD45RA negative) and terminally differentiated (TD CD27 negative CD45RA+) (**Figure 1**) [6].

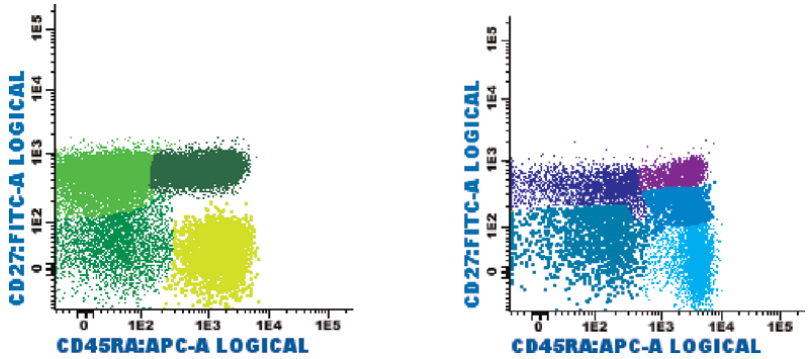

Figure 1.
T-cell subsets based on the expression of CD27 x CD45RA: TCD4+ naïve (dark green), central memory/ transitional memory (CM/TM; bright green), effector memory (EM; green) and terminally differentiated (TD; light green). TCD8+ naïve (purple), CM/TM (dark blue), EM (pale blue), and TD (turquoise). Some effector CD8+ T-cells showed dim CD27 positivity (blue).

The *naive* cells are T-cells yet to confront the enemy.

Effector T cells recognize antigens in lymphoid organs (central) or in peripheral non-lymphoid tissues (terminal) and are activated to perform functions that are responsible for the elimination of microorganisms and, in disease states, tissue damage.

Memory T cells that are generated by T cell activation are long-lived cells with a greater capacity to react against the antigen. After the T cell response subsides, there are many more memory cells of the corresponding clone than immature T cells that existed before the response. These memory cells respond quickly to subsequent encounters with the same antigen and generate new effector cells that eliminate it.

2.4 TCR

T cell receptor is a membrane-bound heterodimer consisting of two polypeptide chains (α/β or γ/δ) and is associated with cytoplasmic protein CD3. During T-cell development, rearrangement of VJ and VDJ (variable (V), diversity (D), joining (J), and constant (C) region) genes of α/β and γ/δ TCR chains provides the molecular basis for the vast diversity of the T-cell recognition repertoire (**Figure 2**) [6].

Only one type of β chain can be expressed on an αβ T cell and therefore all cells in a clonal T cell population express the same unique TCR (**Figure 3**).

Flow cytometry-based analysis of TCR-β variable region families, employing a set of 25 monoclonal antibodies, allows for the characterization and enumeration of approximately 85% of the total human T-cell repertoire and is a useful target to track clonal expansions.

However, TCR-VBeta families analysis is expensive and time-consuming. TCR-Vβ repertoire analysis by flow cytometry is labor-intensive, costly, difficult to interpret, and of limited sensitivity (**Figure 4**). In addition, although next-generation sequencing (NGS) of rearranged TCRs has been an alternative tool with greater resolution and analytic sensitivity, it is highly complex, costly, and not usually available in routine diagnostic practice.

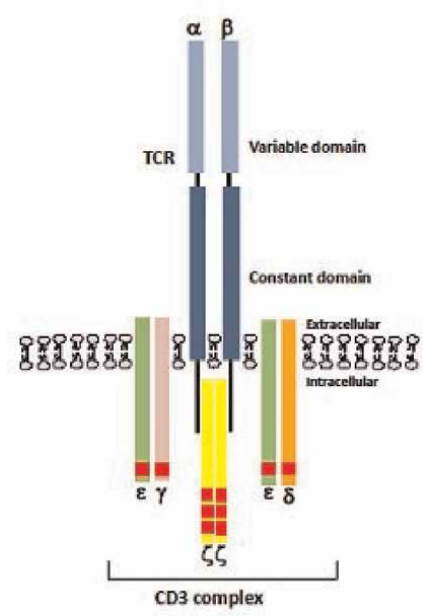

Figure 2.
Schematic representation of a TCR heterodimer consisting of an alpha (α) and beta (β) polypeptide chain, with each polypeptide containing a constant and a variable region.

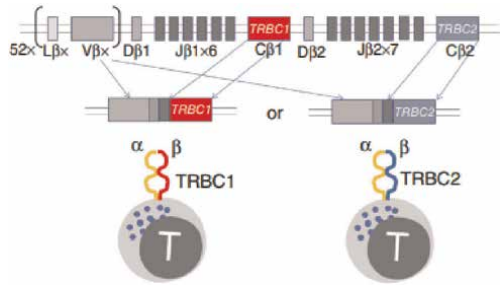

Figure 3.
TCRCBeta1 structure and its V(D)J rearrangements [7].

2.5 When friends become foes

Lymphocytes are the unique cells in the body with clonally expressed antigen receptors, each specific for a different antigenic determinant. Each T lymphocyte clone expresses antigen receptors with a unique specificity, which is different from the specificities of the receptors on other clones.

Thus, there are millions of clones of lymphocytes in the body, allowing the recognition and response to millions of foreign antigens.

The activation of lymphocytes follows a series of sequential steps that begin with the synthesis of new proteins necessary for many of the subsequent changes. The immature cells then begin to proliferate, resulting in an increased size of antigen-specific clones, a process called clonal expansion. In some infections, the number of T cells infected by the microorganism can increase more than 50,000 times. This rapid

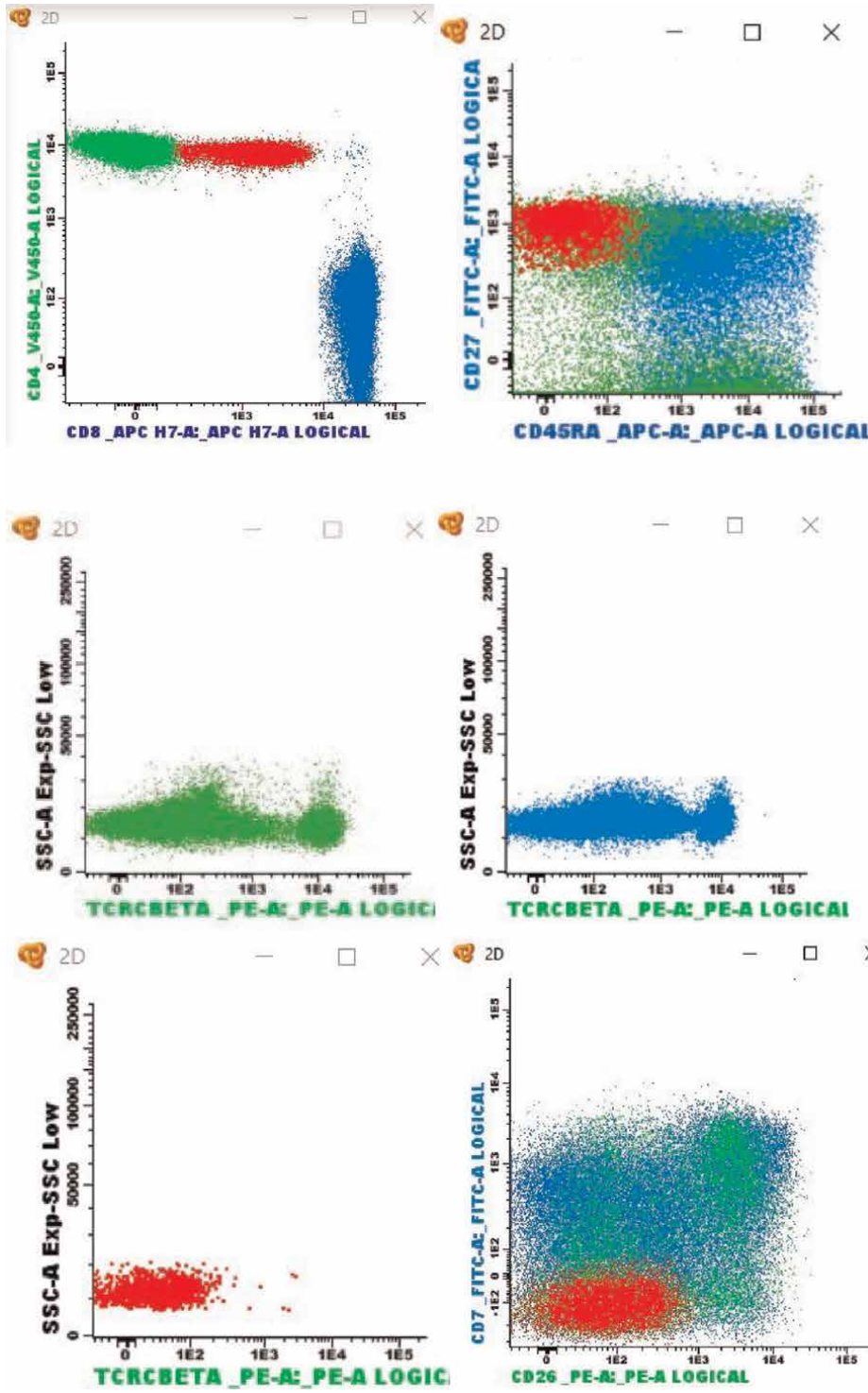

Figure 4.
Flow cytometry dot plot (Infinicyt© software) showing TCD4+ (green) and TCD8+ (blue) normal populations in comparison with monoclonal T-cell population (red: CD4++ CD8 + dim with negative expression of CD7 and TCRBeta1 negative in 100% of cells).

clonal expansion of specific lymphocytes is necessary to keep pace with the ability of microorganisms to rapidly replicate [8, 9].

The rearrangement of antigen receptor genes is a key event in the development of lymphocytes and is responsible for the generation of this diverse repertoire. Each T lymphocyte clone produces an antigen receptor with a unique antigen-binding structure in a genetic process similar to the production of surface immunoglobulins on B lymphocytes. The ability to generate these extremely diverse repertoires does not require an equally large number of genes of different antigen receptors; otherwise, much of the human genome would be dedicated to encoding a large number of TCR molecules.

The genes that encode the different antigen receptors of B and T lymphocytes are generated by the rearrangement, in each lymphocyte, of different gene segments of the variable region (V) with gene segments of diversity (D) and junction (J). This specialized process of rearranging genes at specific locations is called V(D)J recombination.

In αβ T cells, the TCR β chain is the first to be rearranged.

With such a complex genetic mechanism involved in the development of the TCR, it is no wonder that something eventually goes wrong.

Because the Ig and TCR genes are sites of multiple DNA recombination events in B and T cells, and because these sites become active for transcription after recombination, genes from other *loci* can be abnormally translocated to these *loci* and, as a result, may be abnormally transcribed. In B and T lymphocyte tumors, oncogenes are often translocated to the Ig or TCR gene *loci*. These chromosomal translocations are often accompanied by an accentuated transcription of oncogenes and are one of the factors that promote the development of lymphoid tumors [8].

2.6 Recognizing clonal T-cells

Most of the cases of T-cell diseases are easily spotted because of a subpopulation prevalence (TCD4+, TCD8+, T double-positive, or T double-negative). If there is no obvious dominance of a particular T-cell subtype, phenotypic aberrations, most commonly the dim expression of CD3 and dim/negative expression of CD7, maybe the clues.

2.7 Combining the CD45RA and CD27 strategy with TCRCBeta1: separating wheat from chaff

The CD27 and CD45RA phenotypic profile strategy associated with clonality assessment is able to sort out the normal populations from the clonal populations, especially since all clonal populations usually show additional phenotypic aberrancies.

TCRCBeta1, as a strategy to assess T-cell clonality by the addition of a single anti-TRBC1 antibody to a diagnostic flow cytometry T-cell panel, was introduced in 2020 [1, 7, 10–12].

Clonality detection using TRBC1 should always be performed with multiple other T-cell antigens, sorting out different T-cell subsets and separating neoplastic from benign T-cells. Eight to ten-color flow cytometry panels based on previously published data should be used in immunophenotyping.

Thresholds for percentages of TRBC1-positive events greater than 85% or less than 15% might be defined as clonality in current literature.

In our laboratory (data not published), to study CD27 and CD45RA phenotypic profile strategy associated with clonality assessment, TCRCBeta1 (clone JOVI-1) was added to EuroFlow's CLPD-T Tube 2 [13] (CD27, CD45RA, CD8, CD16 and CD56, CD4, CD3, CD45) in the PE channel. The combination of CD27 and CD45RA with TCRCBeta1 proved to be an excellent tool for identifying abnormal T-cell populations, even in small percentages (**Figure 5**).

2.8 Phenotypic features of T-cell chronic lymphoproliferative disorders (T-CLPD)

T-CLPD diagnosis is far from simple; almost 20 different entities are described in 2022 WHO's edition (**Table 1**) [15]. According to the type of population restriction,

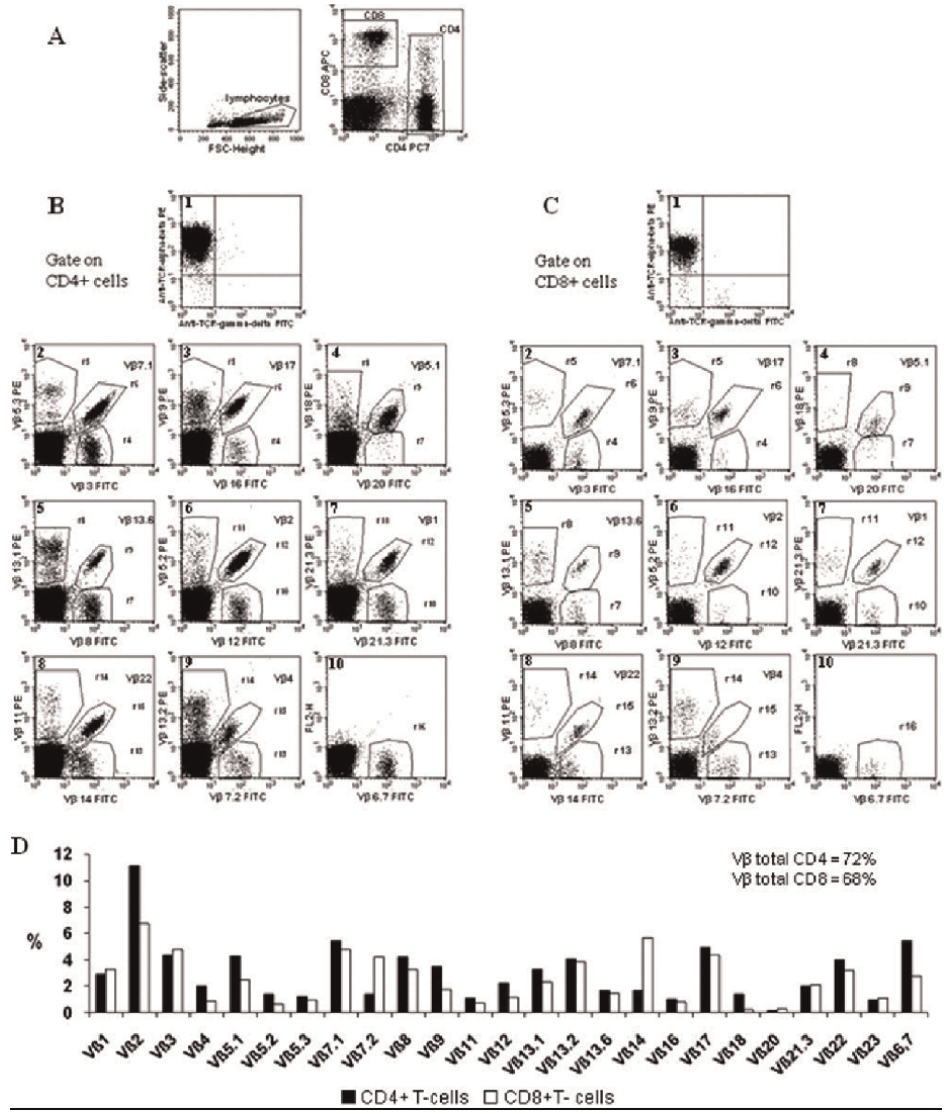

Figure 5.
TCR-VBeta family repertoire (from cytometry part a 75A: 743751, 2009) [14].

4th edition	5th edition
T-lymphoblastic leukemia/lymphoma, NOS	Unchanged
Early T-precursor lymphoblastic leukemia/lymphoma	Unchanged
Adult T-cell leukemia/lymphoma	Unchanged
Sezary syndrome	Unchanged
Primary cutaneous CD4-positive small or medium T-cell lymphoproliferative disorder	Unchanged
Primary cutaneous acral CD8-positive T-cell lymphoma	Primary cutaneous acral CD8-positive lymphoproliferative disorder
Primary cutaneous CD30-positive T-cell lymphoproliferative disorder: Lymphomatoid papulosis	Unchanged
Primary cutaneous CD30-positive T-cell lymphoproliferative disorder: Primary cutaneous anaplastic large cell lymphoma	Unchanged
Subcutaneous panniculitis-like T-cell lymphoma	Unchanged
Primary cutaneous gamma/delta T-cell lymphoma	Unchanged
Primary cutaneous CD8-positive aggressive epidermotropic cytotoxic T-cell lymphoma	Unchanged
Indolent T-cell lymphoproliferative disorder of the gastrointestinal tract	Indolent T-cell lymphoma of the gastrointestinal tract
Enteropathy-associated T-cell lymphoma	Unchanged
Monomorphic epitheliotropic intestinal T-cell lymphoma	Unchanged
Intestinal T-cell lymphoma, NOS	Unchanged
Hepatosplenic T-cell lymphoma	Unchanged
Angioimmunoblastic T-cell lymphoma	Nodal TFH cell lymphoma, angioimmunoblastic-type
Follicular T-cell lymphoma	Nodal TFH cell lymphoma, follicular-type
Nodal peripheral T-cell lymphoma with TFH phenotype	Nodal TFH cell lymphoma, NOS

Table 1.
The 5th edition of the World Health Organization classification of hematolymphoid tumors: Overview of changes and new additions to the classification of T-cell lymphomas.

the expression of CD4 and CD8 can be used to formulate a list of diagnostic possibilities and determine what additional information is required for further classification (**Table 2**) [16].

Thus, the immunophenotypic criteria that have been described as of value for the diagnosis of suspected T-CLPD include: (1) deletion of one or more pan-T molecules (CD7, CD5, CD2, and CD3); (2) expression of a molecule or a combination of two or more molecules not usually expressed by a particular maturational stage, and therefore designated as aberrant (CCR7, CD26, CD27, CD28, CD45RA, and CD45RO); (3) discordant expression of CD3 and TCR on the membrane, and (4) co-expression of CD4 and CD8 and/or the absence of expression of both molecules in an important proportion of T cells.

Such criteria, although useful in daily laboratory routine, are fallible, since a clear immunophenotype impairment has only been found in 60–70% of T-CLPD.

Disease entities	Distinguishing phenotypic features	Additional diagnostic information
CD4+ CD8−		
CTCL / Sézary syndrome	CD7(−) CD26(−) CD23+/−	Characteristic morphology and clinical presentation. HTLV-1(−)
*T-PLL	Usually lacks significant phenotypic aberrancy	80% t(14;14)(q11;q32) or inv.(14) (q11; q32). TCL1 expression
*Adult T-cell leukemia/lymphoma	CD7(−) CD25+ (uniform bright)	HTLV-1+ Endemic Japan and Caribbean
Anaplastic large cell lymphoma	Loss of many pan–T-cell antigens Strong uniform CD30+	Anaplastic morphology *ALK* gene rearrangement
Angioimmunoblastic	Aberrant phenotype. CK10+/−	Characteristic morphology
*Peripheral CL, NOS	Variable phenotype, often aberrant loss of CD5 and/or CD7	Diagnosis by exclusion of other distinct disease entities
CD4− CD8+		
T-cell Large granular lymphocyte leukemia.	Frequent aberrant expression CD5 and/or CD7. Positive expression of NK markers	LGL morphology. Indolent course, associated with cytopenias
Subcutaneous panniculitis-like TCL	Usually only focal CD56, EBV(−), TCR α/β + Perforin+	Must be distinguished from lupus profundus
Hepatosplenic TCL	CD5(−) CD7+ CD16+/− CD56+ CD57(−) TIA-1+ Perforin(−) May be double-negative.	Often TCR γ/δ but may be TCR α/β, EBV (−). Frequent isochromosome 7q. Aggressive clinical course.

*+ positive; (−) negative; +/− heterogeneous expression. *may be double-positive. CTCL, cutaneous T-cell lymphoma; TCL, T-cell lymphoma; NOS, not otherwise specified; T-PLL, T-cell prolymphocytic Leukemia.*

Table 2.
Flow cytometric approach to the diagnosis and classification of TCD4+ and TCD8+ lymphoid neoplasms (adapted from flow cytometric immunophenotyping for hematologic neoplasms, 16).

The immunophenotypic profile of the neoplastic lymphoid T cell is already relatively well characterized in some entities, such as leukemia, chronic T-cell prolymphocytic leukemia, adult T-cell lymphoma/leukemia, and Tgamma/delta+ hepatosplenic lymphoma. In others, such as in angioimmunoblastic T lymphoma, the immunophenotypic is heterogeneous and complex, resulting from intra-tumoral heterogeneity [17].

3. Closing remarks: narrowing down diagnostic hypothesis

What we learned so far

- Due to multiple DNA recombination events and highly active transcription, TCR gene *loci* are sites prone to the development of lymphoid tumors;
- the majority of T-CLPD derive from TCR αβ;
- the β-chain of TCR αβ structure has a variant and a constant region, and there are only two genes associated with the β-chain constant region: TCRBC1 and TCRBC2;

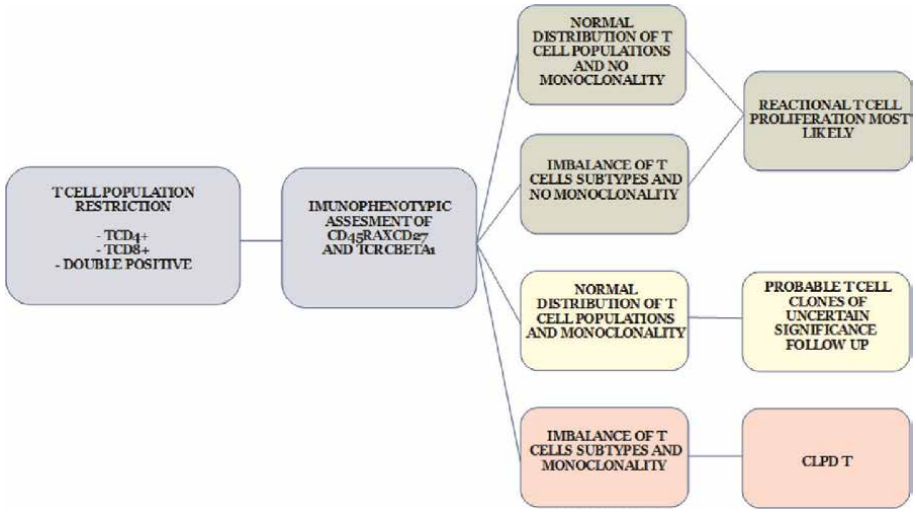

Figure 6.
Algorithm strategy for flow cytometric immunophenotyping for screening and classification of T-CLPD.

- assessment of TCRBC1 by flow cytometry is a fast and easy method for establishing T-cell clonality in TCD4+, TCD8+, and double-positive T-CLPDs;

- phenotypic evaluation of CD27 and CD45RA provides a normal background to sort out an abnormal T-cell population.

Based on all this information, flow cytometry facilities may develop a strategy for performing a CLPD-T diagnosis (or at least a diagnostic hypothesis) (**Figure 6**).

Once the T-CLPD is established, further flow cytometry evaluation is needed to determine any additional phenotypic features, such as lack of expression of T-cell markers, absence of co-stimulatory molecules, stage of maturation arrest, and expression of NK antigens [18].

With all the information in hand, clinical presentation (leukemic, extranodal, and cutaneous) and pathology will conclude the diagnosis.

4. Conclusions

The TCRCBeta1 marker is not only easily implemented in routine immunophenotyping but is also faster and much cheaper than the analysis of TCR-VBeta families either by PCR or flow cytometry.

The incorporation of a future TCRCBeta2 in the panel will refine and consolidate the T Cell clonality assessment and provide an almost definitive threshold between reactive and pathological T cells.

DOI: http://dx.doi.org/10.5772/intechopen.1004631

Author details

Maria Daniela Holthausen Perico* and Renata Kalfeltz
HEMOSC – Hematology and Hemotherapy Center of Santa Catarina, Laboratory Division – Cell Markers Laboratory, Florianopolis, SC, Brazil

*Address all correspondence to: mdanielahemosc@gmail.com

IntechOpen

References

[1] Shi M, Jevremovic D, Otteson GE, Timm MM, Olteanu H, Horna P. Single antibody detection of T-cell receptor αβ Clonality by flow cytometry rapidly identifies mature T-cell neoplasms and monotypic small CD8-positive subsets of uncertain significance. Cytometry Part B (Clinical Cytometry). 2020;**98B**:99-107

[2] Appay V, van Lier RAW, Sallusto F, Roederer M. Phenotype and function of human T lymphocyte subsets: Consensus and issues. Cytometry Part A. 2008;**73A**: 975-983

[3] Purnama C, Camous X, Larbi A. An overview of T cell subsets and their potential use as markers of immunological aging. International Trends in Immunity. 2013;**1**(4):21-32

[4] Mahnke YD, Brodie TM, Sallusto F, Roederer M, Lugli E. The who's who of T-cell differentiation: Human memory T-cell subsets. European Journal of Immunology. 2013;**43**:2797-2809. DOI: 10.1002/eji.201343751

[5] Bretschneider I, Clemente MJ, Meisel C, Guerreiro M, Streitz M, Hopfenmuller W, et al. Discrimination of T-cell subsets and T-cell receptor repertoire distribution. Immunologic Research. Jan 2014;**58**(1):20-27. DOI: 10.1007/s12026-013-8473-0

[6] van der Burg M, Kalina T, Perez-Andres M, Vlkova M, Lopez-Granados E, Blanco E, et al. The EuroFlow PID orientation tube for flow Cytometric diagnostic screening of primary Immunodeficiencies of the lymphoid system. Frontiers in Immunology. March 2019;**10**(246):1-11. DOI: 10.3389/fimmu.2019.00246

[7] Maciocia PM, Wawrzyniecka PA, et al. Targeting the T cell receptor β-chain constant region for immunotherapy of T cell malignancies. Nature Medicine. 2017;**23**:1416–1423. DOI: 10.1038/nm.4444

[8] Abbas AK, Lichtman AH, Pillai S. Cellular and Molecular Immunology. 8th ed. Philadelphia: Elsevier Saunders; 2015

[9] Delves PJ, Roitt IM. Advances in immunology. The immune system. The New-England Journal of Medicine. 2000;**343**(1):37-49. DOI: 10.1056/NEJM200007063430107

[10] Shi M, Olteanu H, Jevremovic D, He R, Viswanatha D, Corley H, et al. T-cell clones of uncertain significance are highly prevalent and show close resemblance to T-cell large granular lymphocytic leukemia. Implications for laboratory diagnostics. Modern Pathology. 2020;**33**:2046-2057. DOI: 10.1038/s41379-020-0568-2

[11] Horna P, Shi M, Jevremovic D, Craig FE, Comfere NI, Olteanu H. Utility of T-cell receptor β constant Chain-1 expression in the diagnosis of peripheral blood involvement by cutaneous T-cell lymphoma. The Journal of Investigative Dermatology. Apr 2021;**141**(4):821-829. e2. DOI: 10.1016/j.jid.2020.09.011

[12] Illingworth A, Wong A, Devitt K, Li W, Furtado FM, Oak J, et al. ICCS Quality and Standards Committee TRBC1 Validation - Practical Guidance. 8 Aug 2023:1-19. Reviewed and approved by: Robert Durso, Regeneron Pharmaceuticals; Lorraine Liu, Vancouver Children's Hospital; David Ng, University of Utah/ARUP Laboratories; Nina Rolf, Vancouver Children's Hospital

[13] Kalina T, Flores-Montero J, van der Velden VHJ, Martin-Ayuso M, Bottcher S, Ritgen M, et al. JJM van Dongen and a Orfao on behalf of the EuroFlow consortium, EuroFlow standardization of flow cytometer instrument settings and immunophenotyping protocols. Leukemia. 2012;**26**:1986-2010. DOI: 10.1038/leu.2012.122

[14] Salameire D, Le Bris Y, Fabre B, Fauconnier J, Solly F, Pernollet M, et al. Efficient characterization of the TCR repertoire in lymph nodes by flow cytometry. Cytometry Part A. 2009;**75A**: 743751

[15] Alaggio R, Amador C, Anagnostopoulos I, et al. The 5th edition of the World Health Organization classification of Haematolymphoid tumours: Lymphoid neoplasms. Leukemia. 2022;**36**:1720-1748. DOI: 10.1038/s41375-022-01620-2

[16] Craig FE, Foon KA. Flow cytometric immunophenotyping for hematologic neoplasms. Blood. 2008;**111**(8):3941-3967

[17] Lima M, Almeida A, Santos AH, Teixeira MA, Algueró ML, Queirós ML, et al. Immunophenotypic analysis of the TCR-Vbeta repertoire in 98 persistent expansions of CD3(+)/TCR-alphabeta (+) large granular lymphocytes: Utility in assessing clonality and insights into the pathogenesis of the disease. The American Journal of Pathology. 2001; **159**:1861-1868

[18] Jevremovic D, Olteanu H. Flow cytometry applications in the diagnosis of T/NK-cell lymphoproliferative disorders. Cytometry Part B. 2019;**96B**: 99-115

Chapter 4

Follicular Helper T Cells and Autoimmune Diseases

Yang Liu, Yanfang Gao, Shiya Wei and Huiqin Hao

Abstract

Follicular helper T (Tfh) cells can control the antibody affinity maturation and memory by supporting the formation of germinal center (GC) and regulating clonal selection and differentiation of memory and antibody-secreting B cells. Therefore, Tfh cells play an important role in the development of some autoimmune diseases, such as rheumatoid arthritis and autoimmune hepatitis. The generation and function of Tfh cells are determined by T-cell antigen receptor (TCR), co-stimulation, and cytokine signals, together with specific mechanisms. In this part, the specialization, development, and regulation of metabolic and differentiation mechanisms on Tfh cells will be summarized, which is crucial to understanding pathogenesis and informing the development of emerging therapies for autoimmune diseases.

Keywords: follicular helper T cells, germinal center, autoimmune disease, B cell, autoantibodies

1. Introduction

The human immune response can be divided into innate immunity and adaptive immunity. Many types of cells of the myeloid lineage are involved in innate immunity, including dendritic cells (DCs), natural killer (NK) cells, macrophages, monocytes, mast cells, and so on. The adaptive immune response is related to the participation of T and B lymphocytes, which will be activated in a specific manner upon recognizing the antigens. The former can be further differentiated into two basic phenotypes according to the surface marker of cluster of differentiation (CD): cytotoxic T cells, which can be distinguished by the CD8, and T helper cells (Th), which are characterized by CD4. As one subpopulation of $CD4^+$ T lymphocytes, follicular helper T cells (Tfh) are usually but not exclusively found in the lymph nodes and spleen, while the other Th cells are predominantly found in lymphoid tissues and blood [1, 2]. They can provide signals directed to B cells located in the germinal center (GC) to promote differentiation and antibody generation [3]. The Tfh-B-cell interaction occurring in the inter-follicular regions of GC is crucial for inducing immune responses efficiently [4].

Autoimmune disease is a category of disease with a prevalence of 7–9% globally that seriously affects human health and results in appreciable mortality [5]. Our knowledge of the roles of Tfh cells in autoimmune diseases has significantly increased during the past decades, and it has been demonstrated that Tfh cells play critical roles in the pathogenesis of many autoimmune diseases, such as rheumatoid arthritis

(RA), autoimmune hepatitis (AIH), systemic lupus erythematosus (SLE), systemic sclerosis (SSc), Primary Sjogren's Syndrome (pSS), myasthenia gravis (MG), inflammatory bowel disease (IBD), immunoglobin G4-related disease (IgG4-RD), etc. [6, 7]. The tissue damage and organ dysfunction of autoimmune diseases are initiated by immune responses mistakenly targeting an individual's cellular, especially by the autoantibody in the humoral immune response. Help to B cells and the formation of DC provided by Tfh allows the affinity maturation of antibody responses and promotes disease pathogenesis by forming immune complexes to mediate inflammation and tissue damage via activating the complement and effector cells in a tissue-specific manner. Although usually located in secondary lymphoid organs, Tfh cells can be identified in human blood, and their frequency and phenotype are often altered in different autoimmune diseases [8].

Herein we will review the recent progress on the specialization and differentiation of Tfh cells, as well as the significance and functionality of these cells in GC formation and the mutation of high-affinity antibody-producing B cells in this chapter. Moreover, how these subsets of Tfh cells engaged in diverse autoimmunity illnesses will also be explored to provide us with novel targets for therapeutic intervention in these disorders.

2. Specialization and development of Tfh

As the unique subset of T lymphocytes with the ability to migrate into or out of follicles, Tfh cells co-localize with B cells in secondary lymphoid organs (SLOs) and deliver contact-dependent and soluble signals to support the survival and differentiation of the latter [9]. Tfh cells can be found in the T-cell zone (pre-Tfh cells phenotype), at the T-B border, and within the GC (the most mature stage), and express different markers in line with the location [10]. The differentiation of Tfh cells is initiated in the T-cell zone of SLOs when naïve $CD4^+$ T cells encounter activated antigen-presenting dendritic cells (DCs). The transcription and translation level of Bcl6 in early pre-Tfh cells elevates within hours at first cell division and then they rapidly gain expression of the canonical surface markers CXC chemokine receptor (CXCR) 5, programmed death receptor (PD)-1, and inducible costimulator (ICOS). The differentiation of early pre-Tfh cell phenotype is also regulated by the transcription factor achaete-scute homolog (ASCL) 2 in a Bcl6-independent manner. ASCL2 can directly induce the upregulation of CXCR5 and downregulation of C chemokine receptor (CCR7) and P-selectin glycoprotein ligand (PSGL)-1 via binding to the Cxcr5 locus [11]. Thereafter, the Tfh cells produce pro-Tfh cytokines, including IL-6 and IL-21, to promote the release of XC chemokine ligand (CXCL)13 by follicular dendric cells (FDCs) and CXCL12 by CXCL12-producing reticular cells (CRCs) in the B-cell zone. Recognition of CXCR5 by CXCL13 leads to recruiting of the immature Tfh into the B-cell zone and to interacting with bystander B cells, which line the follicular parenchyma. Resembling DCs, bystander B cells express inducible costimulator ligand (ICOSL) constitutively to induce ICOS signaling in Tfh cells, which is crucial for follicular retention of Tfh cells and the occurrence of GC reaction [12]. ICOS signaling in Tfh cells further induces CXCR5 and suppresses CCR7, PSGL-1, and CD62L expression through the Kruppel-like factor (KLF) 2, and promotes the motility of Tfh cells in a PI3K-dependent manner to migrate further into the follicle by responding to CXCL13 more robustly [13]. Bystander B cells also express PD-L1 to provide the suppressive signal to follicular entry by activated T cells, which is why continuous ICOS signaling is necessary to maintain the Tfh cells phenotype and follicular

localization, as well as the recruitment of exclusively ICOS-high expressing cells into the follicle. Both CXCR5 and ICOS signaling promote the homing of the now-mature Tfh cells into the GC. GC Tfh cells express higher levels of surface makers, including CXCR5 and PD-1, and more canonical Tfh cell-associated molecules, such as Bcl6, IL-21, IL-4, and Maf, at the transcript level [14]. The localization of Tfh cells to the GC is guided by Epstein-Barr virus-induced molecule (EBI) 2 and sphingosine-1-phosphate receptor (S1PR) 2, and the former (also known as Gpr183) is downregulated in GC Tfh cells and GC B cells while the latter is upregulated [15]. The spatial segregation of Tfh cells in the follicle may also be regulated by surrounding B cells-associated factors, such as plexin B (PlxnB), which can interact with semaphorin 4C (Sema4C) expressed on GC Tfh cells [16]. At last, interactions between Tfh cells and B cells in the GC result in the maturation of B cells into plasma cells to produce antibodies.

3. Subsets and features of Tfh

Human circulating Tfh (cTfh) cells can be subdivided into three main populations according to the expression of surface makers CXCR3 and CCR6: (1) $CXCR3^{+}CCR6^{-}$ Tfh1 cells, that share properties with Th1 cells by secreting interferon (IFN)-γ; (2) $CXCR3^{-}$ $CCR6^{-}$ Tfh2 cells, that resemble Th2 cells by expressing interleukin (IL)-4; (3) $CXCR3^{-}CCR6^{+}$ Tfh17 cells, which are similar to Th17 cells in the production of IL-17. The capacity of Tfh1 cells to assist B cells is not well-defined, while the cTfh2 and Tfh17 cells are considered the most efficient B-cell helpers by inducing naïve B cells to produce immunoglobulins (Tfh2 cells promoting IgG and IgE production and Tfh17 cells promoting IgG and IgA secretion) and promote isotype switching [17].

Human cTfh cells can be further subdivided based on ICOS, PD-1, and CCR7 expression. $ICOS^{-}$ and/or $PD\text{-}1^{-}$ Tfh cells are considered quiescent subsets, whereas $PD\text{-}1^{+}ICOS+CCR7^{lo}$ subsets are recently activated memory Tfh cells with a high capacity to differentiate B cells into IgG-producing cells. This latter population is also described in human autoimmunity, not being abundant in healthy individuals, and positively correlates with disease activity and serum antibody titers of some autoimmune diseases, indicating that it may be a useful biomarker in screening autoimmune patients [18].

4. Regulation of Tfh cells differentiation

The transcriptional environment also changes during these three stages of Tfh cell differentiation. In its primary stage, the expression of Bcl6 is low and the expression of BATF, IRF1, pSTAT5, and TCF-1 is enhanced. During the second stage, Bcl6 expression remains low while pSTAT3, Tbet, and RORγt are upregulated. In the final stage of maturation, Bcl6 is now highly expressed, while the expression of Tbet, GATA3, RORγt, pSTAT5, and Blimp1 is suppressed.

Furthermore, it is increasingly clear that metabolic regulation plays an important role in the differentiation of Tfh cells, including glycolysis, ATP and fatty acid metabolism.

4.1 Glycolysis

Much available data indicates that Tfh cells are distinguished by higher levels of glycolysis and lower levels of oxidative phosphorylation than other subsets of T cells [19].

The process of regulating the glucose metabolism in Tfh cells is very intricate because too low or too high level of glycolysis is not conducive to the differentiation of Tfh cells. Activation of mTORC1 and mTORC2 caused by ICOS can induce the increase in anabolism, high expression of glucose transporter 1 (Glut1) and the promotion of Tfh cell development, while the expression of Bcl-6 inhibits the expression of glycolysis-related enzymes. It is indicated that glycolysis, which is required during early T-cell activation, is not mandatory during the differentiation process of mature Tfh cells. Moreover, IL-2 elevates the activation of Akt and mTORC1 to result in the conversion of low-glycolytic Tfh cells to high-glycolytic Th1 cells [20].

4.2 ATP synthesis and metabolism

ATP-gated ionotropic P2X7, one of the purinergic receptors, is selectively and abundantly expressed in the plasma membranes of Tfh cells and can mediate apoptosis in these cells. Knockouting of P2rx7 can promote Tfh cells to resist apoptosis, elevate the absolute number of Tfh cells in Peyer's patches, and enhance the GC responses significantly [21].

The A2a adenosine receptor (A2aR) is a stimulatory G protein (Gs) protein-coupled G protein-coupled receptor (GPCR) that increases cyclic adenosine monophosphate (AMP) levels. The loss of A2aR leads to an increase in relative Tfh cell differentiation, while the stimulation of A2aR reduces Tfh and GC B-cell numbers. Furthermore, the hypoxic environment inside GC can induce the production of extracellular adenosine to regulate the differentiation of Tfh cells via A2aR [22].

4.3 Fatty acid metabolism

As the proteins that can catalyze the synthesis of monounsaturated fatty acids from saturated fatty acids (SFAs) localized in the endoplasmic reticulum, stearoyl-CoA desaturases (SCDs) are capable of promoting the differentiation of Tfh [23]. It was indicated that fatty acid metabolism is also involved in the development of Tfh cells. However, the mechanism whereby fatty acid metabolism regulates Tfh differentiation remains unclear.

5. Tfh and autoimmune and autoinflammatory diseases

5.1 Tfh and rheumatoid arthritis

Rheumatoid arthritis (RA) is a prevalent chronic autoimmune disease characterized by synovial inflammation and joint cartilage destruction, resulting in joint deformity and disability [24]. It was found that the proportion of cTfh cells in RA patients was significantly higher than that in healthy people [25] and the Tfh cells are increased in the synovium of patients with RA compared with patients with inflammatory osteoarthritis [26]. The frequencies of circulating $CD4^+CXCR5^+CD40L^+$Tfh cells, a Tfh cell subpopulation, are positively correlated with disease activity score-28 with erythrocyte sedimentation rate (DAS28-ESR), rheumatoid factors (RFs), and anti-cyclic citrullinated peptide (anti-CCP) autoantibodies, which are of great significance in the pathogenesis of RA [27]. In addition, the levels of BCL-6 in Tfh cells and IL-21 in the serum of RA patients are higher than those of healthy people, and the expression of IL-21 was positively correlated with disease activity and serum autoantibodies [28].

Deng's research found that in $CD4^+$ T cells of RA patients, excessive activation of the IL-6-pSTAT3 signal leads to abnormal activation of Tfh cells [29].

5.2 Tfh and autoimmune hepatitis

Autoimmune hepatitis (AIH) is a chronic immune-mediated liver disease, featured by circulating autoantibodies, elevated serum IgG levels, and histologic interface hepatitis [30]. Compared with healthy individuals, AIH patients have an increase in Tfh cells and a decrease in follicular regulatory T (Tfr) cells. Moreover, the number of Tfr cells is negatively correlated with the number of Tfh cells, so the imbalance between Tfr and Tfh cells may lead to excessive production of autoantibodies [31]. Ma et al. found that compared with healthy controls, the number of circulating $CD38^+$, $CD86^+$, or $CD95^+$ B cells, $ICOS^+$, and PD-1^+ Tfh cells significantly increased, and serum IL-21 levels increased. The number of $ICOS^+$ or PD-1^+ Tfh cells is positively correlated with $CD86^+$ and $CD95^+$ B cells, respectively. The number of $CD38^+$ B cells, $ICOS^+$, or PD-1^+ Tfh cells is positively correlated with the patient's serum IgG or IgM concentration, respectively. Therefore, it was indicated that circulating activated Tfh and plasma cells may be associated with hypergammaglobulinemia in the pathogenesis of human AIH [32]. However, there is a study reporting that in the experimental autoimmune hepatitis (EAH) model, Tfh cells were lower than in the healthy control group, while Tfr cells were much higher [33]. Besides, the liver injury of EAH mice can be improved by inhibiting Tfh cells or adjusting the imbalance between Tfr cells and Tfh cells [7, 34].

5.3 Tfh and systemic lupus erythematosus

Systemic lupus erythematosus (SLE) is a chronic systemic autoimmune disease marked by impaired immune tolerance and the production of high-affinity autoantibodies. Abnormal production of type I IFN in SLE patients promotes Tfh cells to secrete IL-21 and IFN-γ by activating STAT4, leading to pathogenic B-cell response [35]. In SLE patients, the increased Tfh cells in SLE patients were mostly T-cell factor-1 (TCF1) negative subsets with weakened function, and the changes in these subgroups can reflect the progression of SLE patients [36]. Compared with healthy people, the proportion of cTfh17 cells in SLE patients is increased [37]. However, Szabó et al. found that the frequency of cTfh17 cells decreased in patients with lupus nephritis, which is related to higher disease activity scores [38]. These different results may be due to the use of different markers to detect cTfh17 cells. The frequency of cTfh2 cells was positively correlated with the level of anti-dsDNA autoantibodies and the frequency of plasma cells. Research has also found that interferon γ (IFN-γ) expressing cTfh cells in the MRL/lpr mice are increased [39]. The decrease of Tfr cells and Tfr/Tfh cell ratio in renal lymph nodes are related to the increase of GC-B cells and the onset of lupus nephritis in a mouse model [40].

5.4 Tfh cells and other autoimmune diseases

Primary Sjogren's Syndrome (pSS) is an autoimmune disease with persistent lymphocyte infiltration in exocrine glands, including salivary and lacrimal glands, leading to Sicca syndrome [41]. The percentage and absolute number of PD-$1^+$$ICOS^+$Tfh cells in pSS patients are significantly higher than those in healthy controls, and the number of PD-$1^+$$ICOS^+$Tfh cells is related to B cells. The PD-$1^+$$ICOS^+$Tfh cells are

positively correlated with autoantibodies, erythrocyte sedimentation rate, IgG, IL-2, IL-4, IL-10, IL-17, IFN-γ, TNF-α, IL-21, and disease activity [42].

Systemic sclerosis (SSc) is a connective tissue disease with high mortality, which is characterized by fibrosis of the skin and internal organs [43]. Tfh cell infiltration can be observed in the skin lesions of SSc patients [44]. Compared with healthy controls, the number of cTfh cells in SSc patients increased, the expression level of BCL-6 in cTfh cells was higher, and the ability to secrete IL-21, induce B-cell differentiation and produce IgG and IgM was enhanced [45, 46]. The levels of cTfh1 and cTfh17 in SSc patients increased, and the proportion of cTfh1 cells was positively correlated with autoantibody titer and IL-21 concentration [45].

Inflammatory bowel disease (IBD) is a non-infectious, chronic inflammatory, multifactorial condition encompassing Crohn's disease, ulcerative colitis (UC), and indeterminate colitis [47]. Studies have found that Tfh cells in intestinal GC of IBD patients increase and Tfr cells decrease. In addition, the levels of BCL-6 and IL-21 in the peripheral blood of IBD patients were significantly higher than those of healthy controls [48].

6. Taking Tfh cells into therapy for autoimmune diseases

Increasingly, evidence has demonstrated that the elevation in the number and function of Tfh cells plays an important role in the occurrence and progress of various autoimmune diseases. The study on the mechanism of Tfh cells in autoimmune diseases not only reveals the pathogenesis of autoimmune diseases but also provides a new idea for targeting Tfh cells to treat autoimmune diseases. In this section, we summarize some drugs and methods for Tfh cells to treat autoimmune diseases.

7. Exhausting B cells

Tfh cells can be reduced or eliminated by exhausting B cells that continuously present antigens to Tfh cells, such as using anti-CD20 antibodies. By detecting Tfh cells in the spleen of patients with immune thrombocytopenia treated with rituximab, it was found that all $CD19^+$ B cells in the spleen were missing, and the mature Tfh cells were eliminated, which indicated that B cells might be necessary to maintain Tfh cells in secondary lymphoid organs [49].

7.1 Regulate Tfh cell differentiation

The differentiation of Tfh cells is mainly regulated by the signals of costimulatory molecules and cytokine receptors. CD28, CD40L, ICOS and OX40 are necessary to produce Tfh cells and their auxiliary B cells [3]. By blocking this surface co-receptor, the differentiation of Tfh cells can be inhibited, thus treating autoimmune diseases. For example, CTLA4 fusion protein can block CD28 costimulatory signal and inhibit Tfh cell production by binding CD80 and CD86, thus reducing the number of $PD\text{-}1^+$Tfh cells in RA patients [50] and cTfh cells in pSS patients [51].

7.2 Regulate cytokines and cytokine signals

Targeting cytokines is the most direct and successful treatment strategy for autoimmune diseases. The function of Tfh cells can be regulated by blocking cytokines or cytokine downstream pathways, such as inhibiting IL-6 or Janus kinases (JAKs) [52].

7.3 Regulate intestinal microecology

Intestinal probiotics may reduce the number of pathogenic microorganisms in the intestine and the differentiation of related autoreactive Tfh cells, thus inhibiting the development of autoimmune diseases. Transplantation of fecal microflora can alleviate liver injury in EAH mice by regulating the balance between Tfr cells and Tfh cells [7].

8. Challenge, bottleneck and prospect

Although targeting Tfh cells has shown enormous therapeutic potential in animal models of several autoimmune diseases, more rigorously designed studies are still needed to ensure the biological safety of these therapies. Because Tfh cells play a key role in antibody production, targeting Tfh cells for autoimmune diseases may lead to a decrease of various antibodies, including antibodies that protect us from viruses, bacteria, parasites, and fungi, thus causing serious infections and no response to vaccines [53]. Therefore, more efforts are needed to fully elucidate the role of Tfh cells in autoimmune diseases. We need to further use cutting-edge single-cell technology to analyze the heterogeneity within the Tfh cell population and understand the specificity, plasticity, and stability of specific Tfh cell subsets so that we can better understand which Tfh cells or molecules in Tfh are the most pathogenic in autoimmune diseases. Therefore, fine-tuning the specific subpopulation of Tfh cells or the unique molecules differentially expressed in Tfh cells related to the pathogenesis of autoimmune diseases, and inhibiting pathogenic Tfh cells without affecting the protection against infection, may be the development trend of targeted Tfh cell therapy in the future. In addition, it is also necessary to further implement more precise individualized treatment in targeted Tfh cell therapy, and study how to use Tfh cell characteristics, such as Tfh cell frequency and functional-related molecular testing to stratify patients and guide the treatment plan and prognosis evaluation of targeted immunotherapy.

In this paper, a large number of evidence of Tfh cells and the rapid growth of autoimmune diseases are reviewed, and the incidence and progress of Tfh cells in autoimmune diseases and the research status of targeted therapy are introduced. Although there are still some challenges and bottlenecks, the prospect of using Tfh cells to treat autoimmune diseases is still very optimistic. We hope that through further research and technology development, we can deeply understand the specific mechanism of Tfh cell subsets and specific molecules in autoimmune diseases, and develop safer, more effective, and more accurate treatment strategies for Tfh cells, further transfer targeted therapy from laboratory to clinical application, and bring better health and quality of life to patients.

Acknowledgements

This study was funded by Special Project of Scientific And Technological Cooperation and Exchange in Shanxi Province (Grant Number: 202104041101013, Grant Recipient: Yang Liu); Science and Technology Innovation Talent Team of Shanxi Province (Grant Number: 202204051002033, Grant Recipient: Huiqin Hao); High-Level Key Disciplines Of Traditional Chinese Medicine Project (Grant Number: CZ2023012, Grant Recipient: Huiqin Hao); Integrated Traditional Chinese

and Western Medicine Basic Discipline Construction Project of Shanxi University of Chinese Medicine (Grant Number: 2023XKJS-03, Grant Recipient: Huiqin Hao); Innovative Team of Prevention and treatment of autoimmune hepatitis with integrated Chinese and Western medicine (Grant Number: 2022TD2003, Grant Recipient: Yang Liu).

Author details

Yang Liu[1,2], Yanfang Gao[1,2], Shiya Wei[1,2] and Huiqin Hao[1,2*]

1 College of Basic Medical Sciences, Shanxi University of Chinese Medicine, Jinzhong, PR China

2 Basic Laboratory of Integrated Traditional Chinese and Western Medicine, Shanxi University of Chinese Medicine, Jinzhong, PR China

*Address all correspondence to: hhq@sxtcm.edu.cn

IntechOpen

References

[1] Oliveira YLDC, Oliveira LM, Cirilo TM, et al. T follicular helper cells: Their development and importance in the context of helminthiasis. Clinical Immunology. 2021;**231**:108844

[2] Song W, Craft J. T follicular helper cell heterogeneity: Time, space, and function. Immunological Reviews. 2019;**288**(1):85-96

[3] Vinuesa CG, Linterman MA, Yu D, et al. Follicular helper T cells. Annual Review of Immunology. 2016;**34**:335-368

[4] Gowthaman U, Chodisetti SB, Agrewala JN. T cell help to B cells in germinal centers: Putting the jigsaw together. International Reviews of Immunology. 2010;**29**(4):403-420

[5] Qi J, Liu C, Bai Z, et al. T follicular helper cells and T follicular regulatory cells in autoimmune diseases. Frontiers in Immunology. 2023;**14**:1178792

[6] Pan S, Xiao X, Li T, et al. Definition of follicular helper T cell and cytokines expression in synovial fluid of rheumatoid arthritis. Clinical Rheumatology. 2023;**43**(1):129-135

[7] Liang M, Liwen Z, Jianguo S, et al. Fecal microbiota transplantation controls progression of experimental autoimmune hepatitis in mice by modulating the TFR/TFH immune imbalance and intestinal microbiota composition. Frontiers in Immunology. 2021;**12**:728723

[8] Ueno H, Banchereau J, Vinuesa CG. Pathophysiology of T follicular helper cells in humans and mice. Nature Immunology. 2015;**16**(2):142-152

[9] Shulman Z, Gitlin AD, Targ S, et al. T follicular helper cell dynamics in germinal centers. Science. 2013;**341**(6146):673-677

[10] Ioannidou K, Ndiaye DR, Noto A, et al. In situ characterization of follicular helper CD4 T cells using multiplexed imaging. Frontiers in Immunology. 2021;**11**:607626

[11] Liu X, Chen X, Zhong B, et al. Transcription factor achaete-scutehomologue 2 initiates follicular T-helper-cell development. Nature. 2014;**507**(7493):513-518

[12] Xu H, Li X, Liu D, et al. Follicular T-helper cell recruitment governed by bystander B cells and ICOS-driven motility. Nature. 2013;**496**(7446):523-527

[13] Weber JP, Fuhrmann F, Feist RK, et al. ICOS maintains the T follicular helper cell phenotype by down-regulating Kruppel-like factor 2. The Journal of Experimental Medicine. 2015;**212**:217-233

[14] Suan D, Nguyen A, Moran I, et al. T follicular helper cells have distinct modes of migration and molecular signatures in naive and memory immune responses. Immunity. 2015;**42**:704-718

[15] Moriyama S, Takahashi N, Green JA, et al. Sphingosine-1-phosphatereceptor 2 is critical for follicular helper T cell retention in germinal centers. The Journal of Experimental Medicine. 2014;**211**:1297-1305

[16] Yan H, Wu L, Shih C, et al. Plexin B2 and Semaphorin 4C guide T cell recruitment and function in the germinal center. Cell Reports. 2017;**19**:995-1007

[17] Morita R, Schmitt N, Bentebibel SE, et al. Human blood CXCR5(+)CD4(+)

T cells are counterparts of T follicular cells and contain specific subsets that differentially support antibody secretion. Immunity. 2011;**34**(1):108-121

[18] He J, Tsai LM, Leong YA, et al. Circulating precursor CCR7(lo)PD-1(hi) $CXCR5^+$ $CD4^+$ T cells indicate Tfh cell activity and promote antibody responses upon antigen reexposure. Immunity. 2013;**39**(4):770-781

[19] Zeng H, Cohen S, Guy C, et al. mTORC1 and mTORC2 kinase signaling and glucose metabolism drive follicular helper T cell differentiation. Immunity. 2016;**45**:540-554

[20] Ray JP, Staron MM, Shyer JA, et al. The Interleukin-2-mTORc1kinase Axis defines the signaling, differentiation, and metabolism of T helper 1 and follicular B helper T cells. Immunity. 2015;**43**:690-702

[21] Proietti M, Cornacchione V, Rezzonico Jost T, et al. ATP-gated ionotropic P2X7 receptor controls follicular T helper cell numbers in Peyer's patches to promote host-microbiota mutualism. Immunity. 2014;**41**:789-801

[22] Abbott RK, Thayer M, Labuda J, et al. Germinal center hypoxia potentiates immunoglobulin class switch recombination. Journal of Immunology. 2016;**197**:4014-4020

[23] Son YM, Cheon IS, Goplen NP, et al. Inhibition of stearoyl-CoA desaturases suppresses follicular help T- and germinal center B-cell responses. European Journal of Immunology. 2020;**50**:1067-1077

[24] Li J, Cui J, Wu L, et al. Machine learning and molecular subtype analyses provide insights into PANoptosis-associated genes in rheumatoid arthritis. Arthritis Research & Therapy. 2023;**25**(1):233

[25] Bemani P, Eklund KK, Ali-Hassanzadeh M, et al. Proporion of T follicular helper cells in peripheral blood of rheumatoid arthritis patients: A systematic review and meta-analysis. Expert Review of Clinical Immunology. 2021;**17**(6):667-680

[26] Weyand CM, Goronzy JJ. The immunology of rheumatoid arthritis. Nature Immunology. 2021;**22**(1):10-18

[27] Xu T, Yan T, Li F, et al. The role of different circulating T follicular helper cell markers in rheumatoid arthritis. Journal of Interferon & Cytokine Research. 2022;**42**(3):108-117

[28] Dam EM, Maier AC, Hocking AM, et al. Increased binding of specificity protein 1 to the IL21R promoter in B cells results in enhanced B cell responses in rheumatoid arthritis. Frontiers in Immunology. 1978;**2018**:9

[29] Deng J, Fan C, Gao X, et al. Signal transducer and activator of transcription 3 hyperactivation associates with follicular helper T cell differentiation and disease activity in rheumatoid arthritis. Frontiers in Immunology. 2018;**9**:1226

[30] European Association for the Study of the Liver. EASL clinical practice guidelines: Autoimmune hepatitis. Journal of Hepatology. 2015;**63**(4):971-1004

[31] Liang M, Liwen Z, Juan D, et al. Dysregulated TFR and TFH cells correlate with B-cell differentiation and antibody production in autoimmune hepatitis. Journal of Cellular and Molecular Medicine. 2020;**24**(7):3948-3957

[32] Ma L, Qin J, Ji H, et al. Tfh and plasma cells are correlated with hypergammaglobulinaemia in patients

with autoimmune hepatitis. Liver International. 2014;**34**(3):405-415

[33] Ma L, Zhang LW, Zhuang Y, et al. Exploration the significance of Tfh and related molecules on C57BL/6 mice model of experimental autoimmune hepatitis. Journal of Microbiology, Immunology, and Infection. 2021;**54**(2):221-227

[34] Ma L, Zhang L, Zhuang Y, et al. Lactobacillus improves the effects of prednisone on autoimmune hepatitis via gut microbiota-mediated follicular helper T cells. Cell Communication and Signaling: CCS. 2022;**20**(1):83

[35] Dong X, Antao OQ, Song W, et al. Type I interferon-activated STAT4 regulation of follicular helper T cell-dependent cytokine and immunoglobulin production in lupus. Arthritis & Rhematology. 2021;**73**(3):478-489

[36] Zeng X, Zheng M, Liu T, et al. Changes in the expression of T-cell factor-1 in follicular helper T cells reflect the condition of systemic lupus erythematosus patients. International Immunopharmacology. 2022;**108**:108877

[37] Mao M, Xu S, Lin L, et al. Impact of corticosteroids on the proportions of circulating Tfh cell subsets in patients with systemic lupus erythematous. Frontiers in Medicine. 2022;**9**:949334

[38] Szabó K, Jámbor I, Pázmándi K, et al. Altered circulating follicular T helper cell subsets and follicular T regulatory cells are indicators of a derailed B cell response in lupus, which could Be modified by targeting IL-21R. International Journal of Molecular Sciences. 2022;**23**(20):12209

[39] Jin X, Chen J, Wu J, et al. Aberrant expansion of follicular helper T cell subsets in patients with systemic lupus erythematosus. Frontiers in Immunology. 2022;**13**:928359

[40] Kumar P, Balakrishnan S, Surendra Lele S, et al. Restoration of follicular T regulatory/helper cell balance by OX40L-JAG1 cotreatment suppresses lupus nephritis in NZBWF1/j mice. Journal of Immunology. 2022;**208**(11):2467-2481

[41] Bautista-Vargas M, Vivas AJ, Tobón GJ. Minor salivary gland biopsy: Its role in the classification and prognosis of Sjögren's syndrome. Autoimmunity Reviews. 2020;**19**(12):102690

[42] Wang Y, Guo H, Liang Z, et al. Sirolimus therapy restores the PD-1+ICOS+Tfh:CD45RA-Foxp3high activated Tfr cell balance in primary Sjögren's syndrome. Molecular Immunology. 2022;**147**:90-100

[43] Denton CP, Khanna D. Systemic s clerosis. Lancet. 2017;**390**(10103): 1685-1699

[44] Taylor DK, Mittereder N, Kuta E, et al. T follicular helper-like cells contribute to skin fibrosis. Science Translational Medicine. 2018;**10**(431):eaaf5307

[45] Ly NTM, Ueda-Hayakawa I, Nguyen CTH, et al. Imbalance toward TFH 1 cells playing a role in aberrant B cell differentiation in systemic sclerosis. Rheumatology (Oxford). 2021;**60**(3):1553-1562

[46] Ricard L, Jachiet V, Malard F, et al. Circulating follicular helper T cells are increased in systemic sclerosis and promote plasmablast differentiation through the IL-21 pathway which can be inhibited by ruxolitinib. Annals of the Rheumatic Diseases. 2019;**78**(4):539-550

[47] Zhang ZM, Lin ZL, He BX, et al. Epidemiological analysis reveals a surge

in inflammatory bowel disease among children and adolescents: A global, regional, and national perspective from 1990 to 2019 - insights from the China study. Journal of Global Health. 2023;**13**:04174

[48] Yang Y, Lv X, Zhan L, et al. Case report: IL-21 and Bcl-6 regulate the proliferation and secretion of Tfh and Tfr cells in the intestinal germinal center of patients with inflammatory Bowel disease. Frontiers in Pharmacology. 2021;**11**:587445

[49] Audia S, Rossato M, Trad M, et al. B cell depleting therapy regulates splenic and circulating T follicular helper cells in immune thrombocytopenia. Journal of Autoimmunity. 2017;**77**:89-95

[50] Aldridge J, Andersson K, Gjertsson I, et al. Blood PD-1+TFh and CTLA-4+CD4+ T cells predict remission after CTLA-4Ig treatment in early rheumatoid arthritis. Rheumatology (Oxford). 2022;**61**(3):1233-1242

[51] Verstappen GM, Meiners PM, Corneth OBJ, et al. Attenuation of follicular helper T cell-dependent B cell hyperactivity by abatacept treatment in primary Sjögren's syndrome. Arthritis & Rhematology. 2017;**69**(9):1850-1861

[52] Deng J, Wei Y, Fonseca VR, et al. T follicular helper cells and Tfollicular regulatory cells in rheumatic diseases. Nature Reviews Rheumatology. 2019;**15**(8):475-490

[53] Crotty S. T follicular helper cell biology: A decade of discovery and diseases. Immunity. 2019;**50**(5):1132-1148

Chapter 5

Tissue-Resident Memory T Cells in Skin Barriers

Ling Chen and Zhu Shen

Abstract

The skin is the largest defense organ and immune organ. Establishing immune memory in the skin is a key component of the acquired immune response. The skin harbors several subpopulations of memory T cells in the skin, including tissue resident memory T cells (TRMs). TRMs have a unique transcriptional profile, with the most significant features of long-term survival (lifespan) and long-term residence in skin lesions. Under physiological conditions, TRMs can respond quickly to the challenge of pathogen infection. However, increasing evidence supports their role in the recurrence of chronic inflammatory skin diseases under uncontrolled conditions. Elucidating the characteristics of skin TRMs will help to provide promising strategies for reducing the frequency and severity of skin inflammation recurrence. In this chapter, we plan to discuss the latest consensus on the biology of TRMs, and share our views on the roles of TRMs in the recurrence of inflammatory skin diseases.

Keywords: tissue-resident memory T cells, skin barrier, immunological memory, disease recurrence, skin inflammation, TRMs

1. Introduction

The body is covered by barrier tissues, such as the skin. Pathogens stimulate dendritic cells (DCs) below the skin through these barrier tissues. DC captures incoming antigen(s) and then migrates to local draining lymph nodes, presenting the antigen to naive T cells. Once activated, naive T cells proliferate and transform into effector T cells, migrating to the B cell region or inflammatory site. A small fraction of activated T cells differentiates into memory T cell precursors. Based on effector function, proliferation ability, and migration potential, these precursor memory cells ultimately develop into distinct subgroups of memory T cells, including tissue resident memory T cells (TRMs) [1].

TRMs are one of the main executors of immunological memory. They do not express CD62L or CCR7, but constantly express receptors such as CD69, CD103, and CD49a, exhibiting unique characteristics of long-term survival and low migration in peripheral tissues. The specific expression pattern of these receptors is largely determined by the microenvironment of peripheral tissue in which TRMs are located. TRMs are a specific subgroup of memory T cells, and possess a unique transcriptional profile different from central memory T cells or effector memory T cells [2].

Research has demonstrated that skin TRMs can significantly over-express genes related to the acquisition of free fatty acid metabolism, thereby further enhancing their ability to utilize fatty acid oxidation [3], which suggests their outstanding ability to adapt to harsh peripheral tissue environments such as glucose depletion. This is one of the important mechanisms of long-term survival in TRMs. Apart from their longevity, low migration in peripheral tissues is also a significant feature of TRMs. Once TRMs reside in peripheral tissues, they will not easily return to the circulating blood due to other stimuli (also exceptions) [4]. On the one hand, TRMs can provide rapid first-line protective immunity when exposed to exogenous antigens again, and on the other hand, they may also play a key role in the recurrence of chronic inflammatory diseases such as psoriasis and fixed drug eruption.

2. Long-term survival in peripheral tissues

Research has shown that in the absence of antigen stimulation, the survival of TRMs in peripheral tissues is mainly regulated by the local microenvironment. These local microenvironmental factors are related to tissue specificity, e.g., in skin tissue, keratinocytes and fibroblasts may play a key role. They undergo significant changes in chronic inflammatory skin damage, and are the main source of IL-7, IL-15, and TGF that may promote T-cell survival, trafficking, and interactions with keratinocytes [5]. It has been demonstrated that hair follicle expression of IL-15 is required for the homeostasis of CD8+ TRMs, and IL-7 for both CD8+ and CD4+ TRMs in the skin [6].

Multiple interrelated intracellular signaling pathways may be involved in the long-term survival of TRMs in peripheral tissues, including JAK/STAT5, PI3K/Akt, and Notch signaling pathways. Research has shown that after the action of IL-7 and/or IL-15, both long-lived memory T cells and short-lived effector T cells can quickly activate the JAK/STAT5 pathway, but the phosphorylation level of STAT5 in the former is much higher than that in the latter [7]. Moreover, sustained high-level STAT5 activation can significantly enhance the expression of anti-apoptotic protein BCL-2 and the survival time of memory T cells [8]. The PI3K/Akt pathway is more strongly activated in long-lived memory precursor T cells than in short-lived effector cells, and inhibition of this pathway can lead to the restricted formation of CD8 + memory T cells. However, further research has found that sustained PI3K/Akt activation does not enhance the survival of CD8 + memory T cells, but rather inhibits the expression of IL-7 and IL-15 receptors, STAT5 phosphorylation, and BCL-2 expression, indicating that the survival of TRMs may depend on the optimized balance of PI3K/Akt signaling [9].

Research has shown that the Notch signaling pathway can affect the survival of CD4 + memory T cells in circulating blood by regulating Akt phosphorylation and glucose uptake. Moreover, subpopulations of CD8 + CD69 + TRMs from various tissues can highly express Notch-1. The inhibition of Notch signaling can affect the expression of specific genes such as CD103, Tmem37, and Acer2 in TRMs [10, 11]. Tmem37 can stabilize calcium ion channels and voltage-gated ion channels; Acer2 can hydrolyze ceramides into sphingosine and free fatty acid. Sphingosine-1-phosphate (S1P) has been shown to promote cell survival [12]. The uptake of exogenous free fatty acid is crucial for the long-term survival of skin TRMs. Knockout of lipid transport molecules of fatty acid binding protein (FABP4 and FABP5) increased in psoriasis lesions can affect the long-term survival of CD8 + TRM cells in the skin [13, 14]. It can be seen that Notch signaling is of great importance to TRMs.

It has been shown that skin CD8+ TRMs upregulate the genes related to lipid uptake and metabolism (e.g., FABP4/5), and utilize mitochondrial fatty acids oxidation (FAO) to support their long-term survival [15]. That is to say, metabolic reprogramming is closely related to the longevity of TRMs. This is actually not limited to resident cells in peripheral lesions, as the immune cell subpopulations in circulating blood are also regulated by different metabolic patterns [16].

3. Long-term residency in peripheral tissues

TRMs can survive for several months (e.g., in the lungs) to several years (e.g., in the skin) in different peripheral tissues. Research has confirmed that the CD69, CD49a, CD103, CCR7, and CCL27-CCR10 axes are closely related to the long-term residence of TRMs in peripheral tissues.

CD69 is a membrane-bound type II C-lectin receptor. Due to its rapid appearance on the surface of the plasma membrane after stimulation, it has been considered as a classic early marker of lymphocyte activation. However, increasing evidence suggests that the role of CD69 in the immune system is far more complex than currently recognized. The main mechanism by which CD69 resides in the periphery of TRMs is to inhibit the expression and function of S1PR1 (sphingosine 1-phosphate acid receptor type 1) on the cell surface. S1PR1 is a downstream target of Kruppel-like factor 2 (KLF2) and mTOR, and involved in T cell migration from peripheral tissue. Research has shown that CD8 + TRMs lack S1PR1 expression, and forced expression of S1PR1 or loss of CD69 can significantly prevent peripheral residence of TRMs [17, 18]. It is now clear that cytokines (including TGF and Type I IFN) that induce the phenotype of TRMs can increase the expression of CD69 and CD49a, and inhibit the KLF2 pathway [19].

CD49a (integrinα1) participating in collagen IV binding is likely to mediate the residence of TRMs in mucosal tissue and the basement membrane zone. Blocking CD49a can reduce the number of TRMs in the mucosal site [20]. CD103 (ITGAE, Integrin alpha E) is enhanced in inflammatory diseases. The binding of CD103 to E-cadherin enhances intercellular interactions and adhesion. Lymph node homing receptor CCR7 is involved in the migration of T cells in peripheral tissues and has a synergistic effect with S1PR1. Therefore, downregulation of CCR7 expression may be another important mechanism for TRMs to reside in peripheral tissues [21]. CCR10 and its ligand CCL27 are highly specific chemokines in skin tissue. Research has shown that CCL27 increases not only during the inflammatory process but also maintains a high level several weeks after allergen attack. Consistent with the increased expression of CCL27, a large number of CD4 + CCR10 + T cells still exist in the skin lesions of allergic contact dermatitis several weeks after clinical recovery, suggesting that the CCL27-CCR10 axis is also an important mechanism for TRM cell retention [22].

4. The plasticity of TRMs

TRMs form a heterogeneous population that provides localized protection against pathogens. They can be further classified by their functional outputs and diversity in phenotypes. These classifications of TRMs are linked to their heterogeneity and plasticity [23]. For example, in the skin, there are both CD69 + CD103 + and CD69 + CD103 - populations, as well as both CD8 + CD49a + and CD8 + CD49a - populations of TRMs.

In vitiligo skin lesions, CD8 + CD49a + TRMs produce IFN-γ, perforin, and granzyme B when stimulated with IL-15. On the contrary, CD8 + CD49a- TRMs in psoriatic lesions mainly produce IL-17 cytokines, exacerbating the local inflammation of this skin disease [24].

Important signals for the plasticity of TRMs in multiple tissues include TGF-b, aryl hydrogen receptor (Ahr) ligands, type I IFN, and cytokine IL-15 [25]. TGF-b supports the differentiation of CD103+ TRMs. However, inflammatory cytokines type I IFN, IL-12, and IL-33 can directly suppress this process [26, 27]. The plasticity of TRMs is an important topic. The plasticity mechanism of TRMs still requires extensive research and exploration. Moreover, there is currently no consensus on whether plasticity is beneficial to the human body or weakens the function of specialized immune cells.

5. TRMs in inflammatory skin diseases

The skin is the largest organ in the body. It contains a large number of T cells, up to 2 x 1010 cells, twice the number of T cells in the blood [28]. Roughly speaking, approximately 50% of skin T cells possess the molecular phenotype characteristics of TRMs, and they have been considered to be related to the recurrence and chronic course of inflammatory diseases [14].

5.1 Psoriasis

Psoriasis is a chronic recurrent skin disease closely related to T cell immune disorders, with plaque-type psoriasis being the most common type. It is characterized by thickened red plaques and silvery scales predominantly occurring on the scalp, trunk, and extensor surfaces. Psoriasis shows substantial negative effects on patient quality of life. The pathophysiology characteristics of psoriasis are abnormal proliferation of keratinocytes and infiltration of immune cells in the dermis and epidermis. It involves the innate and adaptive immune systems, with IL-23–IL-17 axis as the principal [29].

A certain number of T cells are often seen infiltrating the skin lesions, including Th17, Th 22, and Tc17 cells [30, 31]. These cells can secrete various cytokines closely related to the occurrence/development of psoriasis, including IL-17, TNF-α, and IFN-γ. The importance of these cytokines has been demonstrated by the therapeutic effects of biological antibodies that block individual cytokines, including TNF-α, IL-23/IL-12p40, IL-23p19, IL-17A, and IL-17 receptors [32, 33].

As is well known, commonly used immunosuppressive therapy can alleviate psoriasis skin lesions by blocking the aforementioned cytokines, but it cannot further prevent disease recurrence. Moreover, recurrent psoriasis lesions often occur in the areas where the original lesions have subsided, suggesting the possibility of pathological immune memory.

Previous research by the author has shown that the molecular levels of CD69, IL-17A, and VEGFA in the remission area of psoriasis still maintain similar high levels to adjacent recurrent skin lesions, indicating that the remission area of the skin lesion still has a risk of recurrence. Moreover, research showed that the high expression of T cell-related genes (LCK and TRCB1) and inflammatory genes (IL-17, IL-22, and IFNG) still existed in the relief skin lesions after 3 months of relief, indicating that active T cells still reside in it [34]. Further research has confirmed that several months after effective treatment of psoriasis vulgaris with methotrexate, NB-UVB, and

anti-IL-12/23 biological antibodies, CD4 + TRMs secreting IL-17 and CD8 + TRMs secreting IL-22 still existed in the epidermis of the subsided skin lesions. These further confirm that TRMs play a crucial role in the recurrence of psoriasis [31].

It is shown that TRMs in psoriasis lesions exhibit a CD8 + CD69+ / CD4 + CD69+ phenotype, or CD8 + CD103+ / CD4 + CD103- phenotype, or another phenotype. These are determined by the local immune microenvironment and disease progression. If CD49a is used as the marker for classification, as mentioned earlier, TRMs in psoriasis are often CD8 + CD103 + CD49a- subgroup that mainly secretes IL-17A [24, 32]. In general, in psoriasis, CD4 + T cells mainly infiltrate the dermis and almost do not express CD103. CD8 + T cells infiltrating the epidermis are positive for CD103, while most cells in the dermis are CD103-. Therefore, most epidermal T cells are CD8 + CD103 + TRMs. Some CD8 + CD103 + TRMs exist in the papillary layer and subpapillary layer. The number of CD8 + CD103 + TRMs in the epidermis is often correlated with epidermal thickness [35], which supports the role of TRMs in the formation of psoriasis lesions.

Whether it is CD4+ or CD8 + T cells, the IL-17A-secreted subgroup in psoriasis lesions can be collectively referred to as TRM17 [36]. Of course, there are also studies that only refer to the CD8 + IL-17 + subgroup as TRM17 [37]. IL-23 is crucial for maintaining autoimmune inflammation in non-lymphocytic tissues. Research has confirmed that the application of anti-IL-23 receptor antibodies in mice after the remission of primary Candida albicans infection led to the loss of skin TRM17 cells. Moreover, clinical treatment with anti-IL-23 antibodies can significantly reduce TRM117 cells in the skin lesions of psoriasis patients [36]. Correspondingly, compared with other targeted biological agents, anti-IL-23 antibodies have the lowest recurrence rate in clinical treatment of psoriasis. These results demonstrate that locally produced IL-23 promotes *in situ* function of cutaneous TRM17 cells.

5.2 Vitiligo

Vitiligo is a skin disease with acquired depigmentation, and it is currently believed that autoimmunity plays a role in the impaired number/function of melanocytes. According to the distribution of lesions, vitiligo can be divided into three different forms: non-segmental, segmental, and mixed-type vitiligo. Vitiligo is associated with genetic polymorphisms involved in immune response and melanogenesis. Its occurrence also involves environmental factors. The following vitiligo refers to non-segmental type.

The pathophysiology of vitiligo is complex, and more and more evidence suggests that CD8+ TRMs, especially the CD49a + subgroup, are involved in the recurrence of vitiligo [38]. CD8 + CD49a + TRMs in vitiligo have been demonstrated to constitutively express perforin and granzyme B, thus exhibiting a strong cytotoxic phenotype. Even in the skin perilesional vitiligo, CD8 + TRMs highly express CD69, CD103, and CXCR3, and exhibit increased IFN-γ and TNF- α production and moderate cytotoxicity activity [24, 39].

IL-15 can promote the survival of TRMs in vitiligo. Research has shown that IL-15-deficient mice exhibit impaired TRM cell formation. Targeting IL-15 signaling with antibodies against CD122 (IL-15 receptor subunit) can inhibit the production of IFN-γ by TRMs, even deplete TRMs from skin lesions, and reverse the established mouse vitiligo model [40]. These studies emphasize not only the role of TRMs in vitiligo recurrence but also provide new potential targets for the treatment of vitiligo.

5.3 Fixed drug eruption and SJS/TEN

Fixed drug eruption (FDE) is a well-defined, circular, erythematous, or violaceous plaque that recurs as one or a few lesions always in fixed locations (e.g., the genitals, lips, and trunk) upon ingestion of a medication of the same or similar structure.

It occurs in exactly the same location as the first instance, indicating the existence of immune memory. The predominance of the CD8 + memory T cell population within the epidermis has been demonstrated in FDE lesions, and they can produce IFN-γ and TNF-α. These T cells constitutively express CD69 and CD103. In addition, the rate of production of IFN-γ is much faster than their peripheral counterparts [14]. IL-15 is crucial in the pathogenesis of FDE. It is mainly derived from the lesional keratinocytes. Even without antigen stimulation, it can maintain the survival of CD8 + memory T cells in the epidermis for a long period of time (>4 years) [41].

Unlike FDE, Stevens-Johnson Syndrome (SJS) and Toxic Epidermal Necrolysis (TEN) are at the extremely severe end of drug eruption spectrum, and may endanger the patient's life. Skin TRMs have been recognized to play a crucial role in the pathogenesis of SJS and TEN [42, 43]; However, more direct evidence is currently needed.

The SJS/TEN-like skin adverse reactions caused by immune checkpoint inhibitors (ICIs) also involve the role of TRMs. ICIs target immune checkpoint molecules, such as programmed cell death protein 1 (PD-1), PD ligand 1 (PD-L1), and cytotoxic T-lymphocyte-associated protein-4 (CTLA-4). The corresponding antibodies include: pembrolizumab and nivolumab targeting PD-1; atezolizumab, avelumab, and durvalumab targeting PD-L1; ipilimumab against CTLA-4. ICI-driven activation of the immune system can induce various immune-related adverse events (irAEs). Among them, cutaneous irAEs (cirAEs) are the most common and earliest to occur. The cirAEs are characterized by a wide range of phenotypes, including life-threatening SJS/TEN [44].

This mechanism of cirAEs may involve various immune cells, including T cells. The blockade of PD-1 and CTLA-4 enhances the activity of IL-17, IFN-γ, and IL-2-producing T-cells [45, 46]. In addition, research has shown that nivolumab treatment can upregulate granular enzyme B and IFN-γ in lesions of metastatic melanoma patients [47]. The clinical and histopathological characteristics of SJS/TEN induced by ICI are similar to those caused by other classic drugs. It is speculated that PD-L1 is usually not detected in epidermal keratinocytes, but ICI treatment increases the expression of PD-L1, thereby activating cytotoxic CD8 + T cells and inducing apoptosis in keratinocytes with high expression of PD-L1 [46, 48]. It is undeniable that the mechanism of SJS/TEN induced by ICIs is still unclear. Whether the cytotoxic CD8 + T cells involved fully conform to the characteristics of TRMs, that is, whether these cytotoxic CD8 + T cells have a memory function, needs further clarification. Furthermore, innate immunity clearly plays a crucial role in this process, but research in this area is still insufficient. Clarifying these immunological characteristics and mechanisms helps to ensure that immunotherapy for cancer patients can be carried out as much as possible, as this is more important for patients.

5.4 Other inflammatory skin diseases

More and more evidence suggests that TRMs are also involved in the progression of other inflammatory skin diseases, such as alopecia areata [49], atopic dermatitis [50], and cutaneous lupus erythematosus [51, 52]. Due to space limitations, we will not elaborate further here.

6. Conclusion

Skin TRMs are important guardians of the human body barrier. They play an important role in defending against pathogen invasion. However, in uncontrolled pathological conditions, TRMs have become one of the important causes of the recurrence of inflammatory skin diseases due to their characteristics of long-term survival (longevity) and long-term residence. Further clarification of the mechanisms of TRMs in terms of residence, survival, and activation will be of great help in developing corresponding intervention targets to reduce disease recurrence.

Acknowledgements

Part of the research work involved in this chapter was supported by the National Natural Science Foundation of China (No. 82073444, 82273537).

Conflict of interest

The authors declare no conflict of interest.

Nomenclature

Ahr	Aryl hydrogen receptor
cirAEs	Cutaneous immune-related adverse events
CTLA-4	Cytotoxic T-lymphocyte-associated protein-4
DCs	Ddendritic cells
FABP	Fatty acid binding protein
FAO	Fatty acids oxidation
ICIs	Immune checkpoint inhibitors
irAEs	Immune-related adverse events
PD-1	Programmed cell death protein 1
PD-L1	Programmed cell death protein ligand 1
S1P	Sphingosine-1-phosphate
S1PR1	Sphingosine 1-phosphate acid receptor type 1
SJS	Stevens-Johnson Syndrome
TEN	Toxic Epidermal Necrolysis
TRMs	Tissue-resident memory T cells

Author details

Ling Chen[1] and Zhu Shen[2*]

1 Department of Dermatology, Daping Hospital, Army Medical University, Chongqing, China

2 Department of Dermatology, Guangdong Provincial People's Hospital (Guangdong Academy of Medical Sciences), Southern Medical University, Guangzhou, China

*Address all correspondence to: zhushencq@hotmail.com

IntechOpen

References

[1] Shin H, Iwasaki A. Tissue-resident memory T cells. Immunological Reviews. 2013;**255**(1):165-181. DOI: 10.1111/imr.12087

[2] Mackay LK, Rahimpour A, Ma JZ, Collins N, Stock AT, Hafon ML, et al. The developmental pathway for CD103(+)CD8+ tissue-resident memory T cells of skin. Nature Immunology. 2013;**14**(12):1294-1301. DOI: 10.1038/ni.2744

[3] Frizzell H, Fonseca R, Christo SN, Evrard M, Cruz-Gomez S, Zanluqui NG, et al. Organ-specific isoform selection of fatty acid-binding proteins in tissue-resident lymphocytes. Science Immunology. 2020;**5**(46):eaay9283. DOI: 10.1126/sciimmunol. aay9283

[4] Carbone FR. Unique properties of tissue-resident memory T cells in the lungs: Implications for COVID-19 and other respiratory diseases. Nature Reviews. Immunology. 2023;**23**(5):329-335. DOI: 10.1038/s41577-022-00815-z

[5] Gröne A. Keratinocytes and cytokines. Veterinary Immunology and Immunopathology. 2002;**88**(1-2):1-12. DOI: 10.1016/s0165-2427(02)00136-8

[6] Adachi T, Kobayashi T, Sugihara E, Yamada T, Ikuta K, Pittaluga S, et al. Hair follicle-derived IL-7 and IL-15 mediate skin-resident memory T cell homeostasis and lymphoma. Nature Medicine. 2015;**21**(11):1272-1279. DOI: 10.1038/nm.3962

[7] Riou C, Yassine-Diab B, van Grevenynghe J, Somogyi R, Greller LD, Gagnon D, et al. Convergence of TCR and cytokine signaling leads to FOXO3a phosphorylation and drives the survival of CD4+ central memory T cells. The Journal of Experimental Medicine. 2007;**204**(1):79-91. DOI: 10.1084/jem.20061681

[8] Chetoui N, Boisvert M, Gendron S, Aoudjit F. Interleukin-7 promotes the survival of human CD4+ effector/memory T cells by up-regulating Bcl-2 proteins and activating the JAK/STAT signalling pathway. Immunology. 2010;**130**(3):418-426. DOI: 10.1111/j.1365-2567.2009.03244.x

[9] Hand TW, Cui W, Jung YW, Sefik E, Joshi NS, Chandele A, et al. Differential effects of STAT5 and PI3K/AKT signaling on effector and memory CD8 T-cell survival. Proceedings of the National Academy of Sciences of the United States of America. 2010;**107**(38):16601-16606. DOI: 10.1073/pnas.1003457107

[10] Maekawa Y, Ishifune C, Tsukumo S, Hozumi K, Yagita H, Yasutomo K. Notch controls the survival of memory CD4+ T cells by regulating glucose uptake. Nature Medicine. 2015;**21**(1):55-61. DOI: 10.1038/nm.3758

[11] Wijeyesinghe S, Masopust D. Resident memory T cells are a Notch above the rest. Nature Immunology. 2016;**17**(12):1337-1338. DOI: 10.1038/ni.3617

[12] Liu Z, Yang H, Zhi L, Xue H, Lu Z, Zhao Y, et al. Sphingosine 1-phosphate stimulates insulin secretion and improves cell survival by blocking voltage-dependent K+ channels in β cells. Frontiers in Pharmacology. 2021;**12**(12):683674. DOI: 10.3389/fphar.2021.683674

[13] Pan Y, Tian T, Park CO, Lofftus SY, Mei S, Liu X, et al. Survival of

tissue-resident memory T cells requires exogenous lipid uptake and metabolism. Nature. 2017;**543**(7644):252-256. DOI: 10.1038/nature21379

[14] Chen L, Shen Z. Tissue-resident memory T cells and their biological characteristics in the recurrence of inflammatory skin disorders. Cellular & Molecular Immunology. 2020;**17**(1):64-75. DOI: 10.1038/s41423-019-0291-4

[15] Pan Y, Kupper TS. Metabolic reprogramming and longevity of tissue-resident memory T cells. Frontiers in Immunology. 2018;**18**(9):1347. DOI: 10.3389/fimmu.2018.01347

[16] Peng L, Chen L, Wan J, Liu W, Lou S, Shen Z. Single-cell transcriptomic landscape of immunometabolism reveals intervention candidates of ascorbate and aldarate metabolism, fatty-acid degradation and PUFA metabolism of T-cell subsets in healthy controls, psoriasis and psoriatic arthritis. Frontiers in Immunology. 2023;**10**(14):1179877. DOI: 10.3389/fimmu.2023.1179877

[17] Mackay LK, Braun A, Macleod BL, Collins N, Tebartz C, Bedoui S, et al. Cutting edge: CD69 interference with sphingosine-1-phosphate receptor function regulates peripheral T cell retention. Journal of Immunology. 2015;**194**(5):2059-2063. DOI: 10.4049/jimmunol.1402256

[18] Skon CN, Lee JY, Anderson KG, Masopust D, Hogquist KA, Jameson SC. Transcriptional downregulation of S1pr1 is required for the establishment of resident memory CD8+ T cells. Nature Immunology. 2013;**14**(12):1285-1293. DOI: 10.1038/ni.2745

[19] Zhang N, Bevan MJ. Transforming growth factor-β signaling controls the formation and maintenance of gut-resident memory T cells by regulating migration and retention. Immunity. 2013;**39**(4):687-696. DOI: 10.1016/j.immuni.2013.08.019

[20] Topham DJ, Reilly EC. Tissue-resident memory CD8+ T cells: From phenotype to function. Frontiers in Immunology. 2018;**26**(9):515. DOI: 10.3389/fimmu.2018.00515

[21] Comerford I, Harata-Lee Y, Bunting MD, Gregor C, Kara EE, McColl SR. A myriad of functions and complex regulation of the CCR7/CCL19/CCL21 chemokine axis in the adaptive immune system. Cytokine & Growth Factor Reviews. 2013;**24**(3):269-283. DOI: 10.1016/j.cytogfr.2013.03.001

[22] Moed H, Boorsma DM, Tensen CP, Flier J, Jonker MJ, Stoof TJ, et al. Increased CCL27-CCR10 expression in allergic contact dermatitis: Implications for local skin memory. The Journal of Pathology. 2004;**204**(1):39-46. DOI: 10.1002/path.1619

[23] Marchesini Tovar G, Gallen C, Bergsbaken T. CD8+ tissue-resident memory T cells: Versatile guardians of the tissue. Journal of Immunology. 2024;**212**(3):361-368. DOI: 10.4049/jimmunol.2300399

[24] Cheuk S, Schlums H, Gallais Sérézal I, Martini E, Chiang SC, Marquardt N, et al. CD49a expression defines tissue-resident CD8+ T cells poised for cytotoxic function in human skin. Immunity. 2017;**46**(2):287-300. DOI: 10.1016/j.immuni.2017.01.009

[25] Yang K, Kallies A. Tissue-specific differentiation of CD8+ resident memory T cells. Trends in Immunology. 2021;**42**(10):876-890. DOI: 10.1016/j.it.2021.08.002

[26] Kumar BV, Connors TJ, Farber DL. Human T cell development, localization,

and function throughout life. Immunity. 2018;**48**(2):202-213. DOI: 10.1016/j.immuni.2018.01.007

[27] Yenyuwadee S, Sanchez-Trincado Lopez JL, Shah R, Rosato PC, Boussiotis VA. The evolving role of tissue-resident memory T cells in infections and cancer. Science Advances. 2022;**8**(33):eabo5871. DOI: 10.1126/sciadv.abo5871

[28] Clark RA. Skin-resident T cells: The ups and downs of on site immunity. The Journal of Investigative Dermatology. 2010;**130**(2):362-370. DOI: 10.1038/jid.2009.247

[29] Greb JE, Goldminz AM, Elder JT, Lebwohl MG, Gladman DD, Wu JJ, et al. Psoriasis. Nature Review in Disease Primers. 2016;**24**(2):16082. DOI: 10.1038/nrdp.2016.82

[30] Eysteinsdóttir JH, Sigurgeirsson B, Ólafsson JH, Fridriksson T, Agnarsson BA, Davíðsson S, et al. The role of Th17/Tc17 peripheral blood T cells in psoriasis and their positive therapeutic response. Scandinavian Journal of Immunology. 2013;**78**(6):529-537. DOI: 10.1111/sji.12114

[31] Cheuk S, Wikén M, Blomqvist L, Nylén S, Talme T, Ståhle M, et al. Epidermal Th22 and Tc17 cells form a localized disease memory in clinically healed psoriasis. Journal of Immunology. 2014;**192**(7):3111-3120. DOI: 10.4049/jimmunol.1302313

[32] Tokura Y, Phadungsaksawasdi P, Kurihara K, Fujiyama T, Honda T. Pathophysiology of skin resident memory T cells. Frontiers in Immunology. 2021;**3**(11):618897. DOI: 10.3389/fimmu.2020.618897

[33] Liu Y, Ouyang Y, You W, Liu W, Cheng Y, Mai X, et al. Physiological roles of human interleukin-17 family. Experimental Dermatology. 2024;**33**(1):e14964. DOI: 10.1111/exd.14964

[34] Suárez-Fariñas M, Fuentes-Duculan J, Lowes MA, Krueger JG. Resolved psoriasis lesions retain expression of a subset of disease-related genes. The Journal of Investigative Dermatology. 2011;**131**(2):391-400. DOI: 10.1038/jid.2010.280

[35] Kurihara K, Fujiyama T, Phadungsaksawasdi P, Ito T, Tokura Y. Significance of IL-17A-producing CD8+CD103+ skin resident memory T cells in psoriasis lesion and their possible relationship to clinical course. Journal of Dermatological Science. 2019;**95**(1):21-27. DOI: 10.1016/j.jdermsci.2019.06.002

[36] Whitley SK, Li M, Kashem SW, Hirai T, Igyártó BZ, Knizner K, et al. Local IL-23 is required for proliferation and retention of skin-resident memory TH17 cells. Science Immunology. 2022;**7**(77):eabq3254. DOI: 10.1126/sciimmunol.abq3254

[37] Park SL, Christo SN, Wells AC, Gandolfo LC, Zaid A, Alexandre YO, et al. Divergent molecular networks program functionally distinct CD8+ skin-resident memory T cells. Science. 2023;**382**(6674):1073-1079. DOI: 10.1126/science.adi8885

[38] Shah F, Patel S, Begum R, Dwivedi M. Emerging role of tissue resident memory T cells in vitiligo: From pathogenesis to therapeutics. Autoimmunity Reviews. 2021;**20**(8):102868. DOI: 10.1016/j.autrev.2021.102868

[39] Boniface K, Jacquemin C, Darrigade AS, Dessarthe B, Martins C, Boukhedouni N, et al. Vitiligo skin is

imprinted with resident memory CD8 T cells expressing CXCR3. The Journal of Investigative Dermatology. 2018;**138**(2):355-364. DOI: 10.1016/j.jid.2017.08.038

[40] Richmond JM, Strassner JP, Zapata L Jr, Garg M, Riding RL, Refat MA, et al. Antibody blockade of IL-15 signaling has the potential to durably reverse vitiligo. Science Translational Medicine. 2018;**10**(450):eaam7710. DOI: 10.1126/scitranslmed.aam7710

[41] Mizukawa Y, Yamazaki Y, Shiohara T. In vivo dynamics of intraepidermal CD8+ T cells and CD4+ T cells during the evolution of fixed drug eruption. The British Journal of Dermatology. 2008;**158**(6):1230-1238. DOI: 10.1111/j.1365-2133.2008.08516.x

[42] Schunkert EM, Shah PN, Divito SJ. Skin resident memory T cells may play critical role in delayed-type drug hypersensitivity reactions. Frontiers in Immunology. 2021;**23**(12):654190. DOI: 10.3389/fimmu.2021.654190

[43] Russomanno K, DiLorenzo A, Horeczko J, Deng M, Cardis M, Petronic-Rosic V, et al. Photodistributed toxic epidermal necrolysis in association with lamotrigine and tanning bed exposure. JAAD Case Report. 2021;**2**(14):68-71. DOI: 10.1016/j.jdcr.2021.05.015

[44] Bhardwaj M, Chiu MN, Pilkhwal SS. Adverse cutaneous toxicities by PD-1/PD-L1 immune checkpoint inhibitors: Pathogenesis, treatment, and surveillance. Cutaneous and Ocular Toxicology. 2022;**41**(1):73-90. DOI: 10.1080/15569527.2022.2034842

[45] Li Q, Ngo PT, Egilmez NK. Anti-PD-1 antibody-mediated activation of type 17 T-cells undermines checkpoint blockade therapy. Cancer Immunology, Immunotherapy. 2021;**70**(6):1789-1796. DOI: 10.1007/s00262-020-02795-2

[46] Watanabe T, Yamaguchi Y. Cutaneous manifestations associated with immune checkpoint inhibitors. Frontiers in Immunology. 2023;**20**(14):1071983. DOI: 10.3389/fimmu.2023.1071983

[47] Anegawa H, Otsuka A, Kaku Y, Nonomura Y, Fujisawa A, Endo Y, et al. Upregulation of granzyme B and interferon-γ mRNA in responding lesions by treatment with nivolumab for metastatic melanoma: A case report. Journal of the European Academy of Dermatology and Venereology. 2016;**30**(12):e231-e232. DOI: 10.1111/jdv.13567

[48] Vivar KL, Deschaine M, Messina J, Divine JM, Rabionet A, Patel N, et al. Epidermal programmed cell death-ligand 1 expression in TEN associated with nivolumab therapy. Journal of Cutaneous Pathology. 2017;**44**(4):381-384. DOI: 10.1111/cup

[49] Sasson SC, Gordon CL, Christo SN, Klenerman P, Mackay LK. Local heroes or villains: Tissue-resident memory T cells in human health and disease. Cellular & Molecular Immunology. 2020;**17**(2):113-122. DOI: 10.1038/s41423-019-0359-1

[50] Zheng C, Cao T, Ye C, Zou Y. Neutrophil recruitment by CD4 tissue-resident memory T cells induces chronic recurrent inflammation in atopic dermatitis. Clinical Immunology. 2023;**256**:109805. DOI: 10.1016/j.clim.2023.109805

[51] Zhao Z, Zhu H, Li Q, Liao W, Chen K, Yang M, et al. Skin CD4+ Trm cells distinguish acute cutaneous lupus erythematosus from localized discoid lupus erythematosus/subacute cutaneous lupus erythematosus and other skin diseases. Journal of Autoimmunity.

2022;**128**:102811. DOI: 10.1016/j.jaut.2022.102811

[52] Gu HJ, Song S, Roh JY, Jung Y, Kim HJ. Expression pattern of tissue-resident memory T cells in cutaneous lupus erythematosus. Lupus. 2021;**30**(9):1427-1437. DOI: 10.1177/09612033211017218

Edited by Hilal Arnouk

More than half a century of groundbreaking discoveries in the field of immunology have been made, and no cells have shown more potential than the lymphocytes known as T cells. These little miracles play a crucial role in protecting us from infectious diseases and cancer, as well as predisposing us to certain diseases, such as autoimmunity, when they are dysfunctional or dysregulated. This book aims to foster collaboration between world-class bench scientists and clinicians in the fields of immunology, pathology, and oncology as they come together to highlight the complexity and diversity of T cells as well as their contributions to our immune homeostasis and overall health and to discuss the revolutionary therapeutic applications of T cells in fighting infectious diseases, autoimmunity, and malignant neoplasms.

Published in London, UK

IntechOpen

ISBN 978-0-85466-864-9
9 780854 668649