Changes of cytoskeleton affect T cell biological behaviors

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1. ABSTRACT

T lymphocyte (T cell) is a crucial member in adaptive immunity that protects the organism against foreign pathogens and cancer by producing cytokines and cytotoxic molecules. In T cell, the cytoskeleton is in a stable and dynamic renewed condition and maintains shape and mechanical resistance to deformation. The cytoskeleton changes lead to morphological differences of T cell, and then affect local mechanical properties and cellular behavior, including development, differentiation, polarization, migration or adhesion. In this review, we will discuss the effect of cytoskeleton changes on T cell biological behaviors and its mechanism.

2. INTRODUCTION

T lymphocyte (T cell) is one of a major kind of immune cells in cell-mediated immunity, participating acquired immune response. In the bone marrow, All T cells originate from haematopoietic stem cells, which develop into haematopoietic progenitors (lymphoid progenitor cells). These progenitor cells populate the thymus and expand by cell division to generate a large population of immature thymocytes (1). In the thymus, the earliest thymocytes without CD4+ or CD8+ expression finally mature to single-positive (CD4+CD8- or CD4-CD8+) thymocytes that are then released from the thymus to peripheral tissues. When T cells are stimulated by antigen, native or memory T cells are mature or activated, and then migrate and recruit into the lesions.

As most cells of all domains of life (archaea, bacteria, eukaryotes), T cells have the structure of cytoskeleton. The cytoskeleton is a network of fibers composed of proteins to maintain shape and mechanical resistance to deformation (2). It’s a both stable and dynamic structure that parts of which are constantly destroyed, renewed or newly constructed (2). It’s like all eukaryotic cells that the major components of T cell are microfilaments composed of the protein actin and microtubules composed of the protein tubulin (3). It’s the interaction between cytoskeleton and hundreds of associated proteins including molecular motors, crosslinkers, capping proteins and nucleation promoting factors that make the cytoskeleton complexity and affect local mechanical properties and cellular behavior (4).

In the rat thymus, the Eph family of receptor tyrosine kinases and their ligands, the ephrins drive intrathymic T cell development by regulating
cytoskeleton that Eph A-Fc or ephrin A-Fc fusion proteins to fetal thymus organ cultures interferes with T cell development (5). In pediatric T-cell leukemias, malignant T lymphocytes can be regulated partly by ephrin-B1 through the control of lipid-raft-associated signaling, adhesion, and invasive activity (6). The T cell activation and migration can be negatively influenced by stress hormones via regulation of key cytoskeletal and plasma membrane factors (7). All these findings proposed the relationship between cytoskeleton and T cell activation and function, which may be crucial in normal immune response, immune disorders and T cell-related diseases. In this review, we will discuss the effect of cytoskeleton changes on T cell biological behaviors and its mechanism.

3. CYTOSKELETON INTERACTION IN UROPOD FORMATION OF T CELL

T cell movement consists of a dynamic interplay between attachment at the cell front and detachment at the rear cell edge, as well as traction machinery that pulls the net cell body forward. The moving cell becomes polarizing as adhesion and detachment occurring at opposite cell edges (8,9). This asymmetry develops between two opposite cell edges—the leading edge and the uropod in T lymphocytes. The uropod in T cell is a specialized pseudopod-like projection with motility and recruitment of bystander cells. Several intercellular adhesion molecules (ICAMs) concentrate at the uropod. The uropod formation can be induced by cAMP agonists and chemokines, respectively, and involve myosin-based cytoskeleton (10). The microtubules retract into uropod could induce T cell deformability, leading to facilitating migration through constricted spaces (11). The acquisition of a motile phenotype in T cells lead to the asymmetric redistribution of ganglioside GM3- and GM1-enriched raft domains to the leading edge and to the uropod, which is needed for T cell migration (12).

In the uropod of T cell, the active cathepsin X modifies the beta2 cytoplasmic tail of lymphocyte function associated antigen 1 (LFA-1), which could promote a high affinity conformation of T cell. This interaction is restricted to the uropod and results in the stabilization of this region, promoting LFA-1-mediated cell uropod elongation (13). When LFA-1 links to the cytoskeleton through alpha-actinin-1 and disruption at the level of integrin or actin, T cell spreading and migratory speed is decreased due to a failure of attachment at the leading edge (14). Recent study further found that the affinity LFA-1 to the anterior and posterior membrane was organized by Myosin Ila via Mst1 regulation, resulting in regulating polarization and adhesion during T cell migration (15).

4. ACTIN CYTOSKELETON CHANGES IN T CELL BIOLOGICAL BEHAVIORS

Actin is a globular multi-functional protein and is a major component of cytoskeleton that forms microfilaments. The actin-based cytoskeleton can be linked to extracellular matrix by alpha-dystroglycan, which is essential for normal development and differentiation of T cells (16). The normal T cell responses can be drived by dynamic cytoskeletal remodelling, which is regulated by canonical formin p140mDia1 in mice (17). Recent study further demonstrated in mice that during T-cell-dependent antibody responses and germinal center reaction, the T cell function is regulated by actin-bundling protein L-plastin (18).

After T cell activation, CD44-dependent adhesion of T cells to hyaluronan is regulated by the cross-linking of CD3, interleukin (IL)-2, tumour necrosis factor-alpha, MIP-1beta and interleukin-8, which is manifested by polarization, spreading and co-localization of cell surface CD44 with a rearranged actin cytoskeleton in hyaluronan-bound T cells (19). In particular, IL-2 could induce human T cell adherence to laminin, collagen type IV and fibronectin. Inhibition of T cell adherence and migration apparently involves abrogation of the rearrangement of the T cell actin cytoskeleton (20), which can be induced by galectin-1 (21).

4.1. CXCL12/SDF-1alpha in actin cytoskeleton changes of T cell

In the movement of T cell, tubulin-dependent cellular deformability is not the rate-limiting factor for locomotion, the increase in migratory activity subsequent to colcemid-treatment is due to a secondary phenomenon, most likely the activation of the actin cytoskeleton (22). SDF-1alpha can induce T cell polarization and migration (23). The chemokine CXCL12/stromal cell-derived factor (SDF)-1 induces migration of lymphoid (24). In both murine and human T cells, the migration and adhesion in response to CXCL12/SDF-1alpha can be stimulated by ITK via altering the actin cytoskeleton (25). Two upstream activators of ITK, Src kinases and PI3K can be activated by CXCR4 (25). CXCL12 induces T cell migration, which is promoted by the interaction between T cell specific adapter protein
(TSAd) interacts with Tec kinase ITK (26). Thus, CXCR4 induces Src kinases and PI3K, leading to ITK activation, and then followed by CXCL12/SDF-1alpha stimulation, finally resulting in the migration and adhesion of T cells.

In the upstream of CXCL12/SDF-1alpha, the inhibition of cytoskeleton reorganization prior to stimulation by semaphorin 3F leads to a repulsive effect on thymocyte migration and inhibition of CXCL12- and sphingosine-1-phosphate-induced thymocyte migration in human (27). In addition to CXCL12, the SDF-1alpha-induced T cell interactions with fibronectin and endothelial cells can be inhibited by allicin by down-regulating cytoskeleton rearrangement (28). In downstream of CXCL12/SDF-1alpha, Wiskott-Aldrich syndrome protein, Crk associated substrate, Nck and focal adhesion kinase were all phosphorylated after SDF-1alpha stimulation and then participate SDF-1alpha-induced migration of the human T-cell line Jurkat (29).

4.2. TCR in actin cytoskeleton changes of T cell

The T cell receptor (TCR) is expressed on the surface of T cell that is responsible for recognizing antigens bound to major histocompatibility complex (MHC) molecules (30). In T cell, the reorganisation of the actin cytoskeleton and the formation of the immunological synapse is induced by TCR, which involves Nck, a adapter protein and built of one SH2 domain and three SH3 domains, through a direct binding to components of the TCR/CD3 Complex and a recruiting o the TCR complex via phosphorylated Slp76 (31). In turn, the TCR down-modulation requires efficient reorganization of the actin cytoskeleton as well as functional non-muscle myosin IIA in CD4+ T cell (32). The TCR signaling can be inhibited by disruption of the proper organization of T cell actin cytoskeleton and downregulation of the RhoA pathway in imunosuppressive activity (33).

4.3. Rho family in actin cytoskeleton changes of T cell

During the migration of human effector memory CD4+ T cell driven by TCR, Rho family GTPases appear a differences performance to activate the cytoskeleton that is consistent with the morphological differences in response to distinct recruitment signals (34). Through stimulating Rac to activation, the actin cytoskeleton is remodeled and then T cell responsiveness is regulated, which can be due to DOCK2, a mammalian homolog of Caenorhabditis elegans CED-5 and Drosophila melanogaster Myoblast City (35). Thus, Rho GTPases are involved in the regulation of actin cytoskeleton in T cell.

In the development of human T cell, Rho GTPase Cdc42 is essential that dominant negative Cdc42 induces a decreased thymopoiesis with an increased apoptosis and decreased proliferation (36). It’s both activation of the small GTPase RhoA or transfection with the constitutively active mutants V14RhoA or V12Rac1 that can abolish the alpha4beta1-induced cell polarization but not affect cell spreading (23). However, the increased GTP-RhoA in T cell can’t form an intact cytoskeleton or migrate toward a chemokine gradient, and the T cell infiltration into inflamed compartments is prevented by 3-hydroxy-3-methylglutaryl-CoA reductase inhibitors (37). When Raf is inhibited by sorafenib or shRNA-mediated knockdown of B-Raf, the T cell spreading on the alpha4beta1 integrin ligands vascular cell adhesion molecule 1 (VCAM-1) is inhibited, suggesting the association of alpha4beta1 integrin with the actin cytoskeleton was dependent on B-Raf activity or expression (38).

Moreover, protein kinase Cε (PKCε)-mediated phosphorylation on Thr-7 regulates Rab5a trafficking to the cell leading edge, then leading to Rac1 activation, actin rearrangement, and T-cell motility, suggesting the axis of PKCε-Rab5a-Rac1 in the regulation of cytoskeleton remodeling and T-cell migration (39). In patients with arteriosclerosis obliterans, the migration of CD4+ T cells can be attenuated by microRNA-142-3p through regulating actin cytoskeleton via RAC1 and ROCK2 (40). Thus, in contrary with RhoA, Rac1 can affect T cell migration, even in pathological condition.

4.4. PI3K in actin cytoskeleton changes of T cell

In regulating actin cytoskeleton remodeling after CD43 ligation, several signaling molecules are essencial, including Src kinases, phospholipase C-gamma2, protein kinase C, extracellular-regulated kinase 1/2, p38 and phosphatidylinositol-3 kinase (PI3K) (41). The PI3K can stimulate the serine/threonine kinase Akt in T cell (42). The PI3K/Akt signaling is crucial in T-cell polarization and lamellipodia formation via the re-organization of the actin cytoskeleton (43). In human CD4+ T cell, PI3K signaling is important in maintaining normal morphology and basal motility as well (44). The Akt is also indispensable for migration via cytoskeleton rearrangements in CD4+ T cell (45). During T cell
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transendothelial migration, AKT1/PI3K converge Src to induce cytoskeleton remodeling (46).

5. F-ACTIN CYTOSKELETON

The activation of native T cell leads to an upregulation of membrane receptors, directional secretion of signaling molecules and profound alterations in the cytoskeletal architecture (47), which are mediated by a dynamic rearrangement of not only the microtubules but also F-actin, and lead to T cell polarization (48). These changes of F-actin and microtubules are a prerequisite for efficient cell migration and adhesion (47). During this cytoskeleton rearrangement, the ezrin–radixin–moesin (ERM) proteins, the family of cytoskeletal proteins, make a cross-linking of the F-actin filament network to integral plasma membrane receptors (49). Moesin, a

Figure 1. CXCR4 induces the migration and adhesion of T cells. CXCR4 induces Src kinases and PI3K, leading to ITK activation, and then followed by CXCL12/SDF-1alpha stimulation, finally resulting in the migration and adhesion of T cells.

Figure 2. The rearrangement of microtubules and also F-actin lead to T cell polarization. The activation of native T cell leads to an upregulation of membrane receptors, directional secretion of signaling molecules and profound alterations in the cytoskeletal architecture, which are mediated by a dynamic rearrangement of not only the microtubules but also F-actin, and lead to T cell polarization.
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member of ERM family, is the primary phosphorylated ERM subject playing a role of dynamic regulation in T cell migration (50). The activated ERM cooperated proteins with flotillins to promote efficient chemotaxis of T cells by structuring the uropod of migrating T cells (51). Moreover, through ERM proteins, the effector T cell depolarization can be induced by glucocorticoids, resulting in impeding migration and APC conjugation (52).

In addition, during T cell development and activation, polarized activation of TCR-CD3-stimulated T cells make F-actin-rich membrane protrusions gather Coronin-1 is inequilibrium between the cytosol and the cell cortex (53). The direct F-actin interactions of flotillin-2 and raft/membrane association of flotillin-2 are essential in human T cell uropod formation (54).

In pathological conditions, when gammadelta T cell migrates into the pleural cavity of mice during diverse inflammatory response, F-actin cytoskeleton has reorganized dependent on 5-LO-derived lipid mediator LTB(4) and its receptor BLT1 (55). In T-cell spontaneous cell migration and chemotaxis, F-actin co-localizes with Gem, a member of the small GTP-binding proteins within the Ras superfamily in the presence of SDF-1 and CXCL12 in the condition of HTLV-1 viral infection (56).

6. CONCLUSIONS

In the changes of T cell cytoskeleton, actin cytoskeleton is the major components to be regulated by several signalings, especially involving CXCL12/SDF-1alpha, TCR, Rho family and PI3K. These signalings change T cell cytoskeleton, finally affecting the development, differentiation, polarization, migration or adhesion of T cell. Moreover, these processes are also regulated by different factors. The network of T cell cytoskeleton regulation is complex. However, the key hub of this is still need to investigate in future, which may provide further understanding on T cell performance in normal immune response and in pathological conditions.

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8. REFERENCES


22. Nikolai G, Niggemann B, Werner M, Zänker KS. Colcemid but not taxol modulates the migratory behavior of...
DOI: 10.1016/S0171-2985(99)80050-3

DOI: 10.1002/jcp.20624


DOI: 10.1074/jbc.M312848200

DOI: 10.1371/journal.pone.0009761

DOI: 10.1371/journal.pone.0103405

DOI: 10.1111/j.0019-2805.2004.01841.x

DOI: 10.1016/S0014-4835(02)00823-8


DOI: 10.1186/1478-811X-7-1

DOI: 10.1038/cmi.2011.41

DOI: 10.1016/j.trim.2010.06.009

DOI: 10.4049/jimmunol.1201817

DOI: 10.1016/S1074-7613(03)00169-9

DOI: 10.3324/haematol.2009.006890
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**Abbreviations:** ICAMs, intercellular adhesion molecules; LFA-1, lymphocyte function associated antigen 1; SDF-1, stromal cell-derived factor 1; TSAd, T cell specific adapter protein; TCR, T cell receptor; PI3K: phosphatidylinositol-3 kinase; ERM, ezrin–radixin–moesin

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