Metabolism plays the key roles in Th cells differentiation

A. Hosseinzadeh¹, S. Soukhtehzari², M. Ghaedi³, R. Mansouri¹

¹Department of Immunology, Faculty of Medicine, Shahid Sadoughi University of Medical Sciences, Safaieye, Yazd/Iran; ²Department of Medical Biotechnology, Tarbiat Modarres University, Medical School, Tehran, Iran; ³Department of Immunology, School of Public Health, Tehran University of Medical Sciences, Tehran, Iran.

INTRODUCTION

Immunometabolism has become an attractive field for both immunologists and nutritionists over the last decades. Nutritional research has shown that specific diets can increase the longevity of some patients with autoimmune conditions (1-4). From the immunological point of view, the mechanisms involved are not fully understood. In recent years, immunological research has focused on revealing these mechanisms. Meanwhile, there is the question of whether and how immunometabolism and autoimmunity are linked to each other.

Autoimmunity diseases occur when the immune system attacks self-antigens or when it cannot regulate the intensity of immune responses. The reasons for autoimmunity disorders are not clear enough. The most common theory is that genetic factors along with environmental factors like diet, as one of the most influential factors, play critical roles. Moreover, nutritional elements (whether micro or macro) have well-known effects on the immune system. Excess supply or deficiency of specific metabolites can also have considerable impact on the functions of the immune system (5-7). Among various immune cells, T CD4⁺ cells play a critical role. There is a body of evidence, which states that metabolites have enormous effects on T CD4⁺ cells and alter their functions resulting in autoimmune diseases.

SUMMARY

The increasing rate of autoimmunity in recent decades cannot be related to only genetic instabilities and disorders. Diet can directly influence our health. Studies have shown that there is a relationship between nutritional elements and alteration in the immune system. Among immune cells, the function of T lymphocyte is important in directing immune response. T CD4⁺ cells lead other immune cells to respond to pathogens by secreting cytokines. HIV⁺ patients, who have largely lost their T CD4⁺ cells, are susceptible to opportunistic infections, which do not normally affect healthy people. It seems that the metabolism of T cells is critical for their differentiation and their consequent functions. After activation, T cells need to undergo clonal expansion, which is a high energy-consuming process. Studies have shown that specific metabolites deprivation or their excess supply affects T CD4⁺ cells subsets differentiation. Abnormal induction of subsets of T CD4⁺ cells causes some autoimmunity reactions and hyper-sensitivity as well, which may result from imbalance of diet uptake. In this mini-review, we describe the findings about fatty acids, glucose, amino acids, and vitamins, which are effective in determining the fates of T CD4⁺ cells. These findings may help us uncover the role of diet in autoimmune diseases.

Key words: T helper cells; metabolism; mTOR, autoimmunity.
THE ROLE OF T CD4+ LYMPHOCYTES IN THE IMMUNE SYSTEM

Among acquired immune cells, T lymphocytes play a critical role in proper immune response. T lymphocytes are divided into T helper (T CD4+) and T cytotoxic (T CD8+) with different functions. T CD4+ cells lead other immune cells, i.e., B lymphocytes and monocytes, to respond to pathogens by secreting cytokines. On the other hand, T CD8+ cells kill target cells in a cell to cell manner. T CD8+ cells are important in cellular immune response, while T CD4+ cells functions are mainly mediated indirectly by arranging other immune cells.

T CD4+ cells are classified in T effector subsets including: Th1, Th2, Th17 (Teff), and T regulatory (Treg) subsets. Th1 cells secrete IFN-γ and active monocytes, while Th17 secretes IL-17. These subsets respond to intracellular pathogens. Th2 cells secrete IL-4 and IL-5, which eradicate extracellular pathogens like fungi and parasites. Th2 cells also are involved in hyper-sensitivity reactions. All these subsets protect against widespread pathogens. Treg cells are unique in their function. After a primary immune response, the immune system returns to the pre-activation situation.

T CD4+ cells malfunction may lead either to susceptibility to infections and cancers, or it may cause self-antigen response, which results in autoimmunity. The T CD4+ cells function is clear in some autoimmune diseases like rheumatoid arthritis (RA), multiple sclerosis (MS), inflammatory bowel disease (IBD) and psoriasis, whilst it is under investigation in other diseases. Specifically, studies show that Th17 are involved in RA pathogenesis and Treg and Breg cells are also down-regulated. Thus, in these patients, Treg therapy may be a potential strategy (8).

UNIQUE FEATURES OF T CD4+ CELLS METABOLISM

T CD4+ cells are potentially able to arrange an innate and acquired immune system. They secrete a wide range of cytokines and regulate the switching of the immune response (13, 14). Moreover, T CD4+ cells play key roles in the pathogenesis of some autoimmunities. Therefore, if T CD4+ cells function improperly, they can imperil the body health.

Immune responses are anabolic reactions, the initiation of which is a highly energy-consuming process. Before the activation of T CD4+ cells, they present no activity at resting phase. Their basic metabolism is represented by catabolism (15). During the resting phase, T CD4+ cells obtain their energy through autophagy and lipid oxidation (9, 16). Soon after activation, T CD4+ cells undergo a metabolic reprogramming. They switch catabolic metabolisms to anabolic ones (17). Anabolism is a required process for DNA, protein, and lipid synthesis, all of which are essential molecules for cell growth and proliferation. In fact, T CD4+ cells like cancer cells need to rapidly undergo a clonal expansion. Hence, they both need high amounts of energy in a short time and have to use a new strategy. In 1924, Otto Warburg reported that...
cancer cells utilize glycolysis to supply energy while normal cells exploit oxidative phosphorylation. This specific metabolism is called the Warburg effect, which is used by activated T CD4⁺ cells (18-22). At a first glance, this strategy does not seem logical. However, when cells undergo mass proliferation to clonal growth (like cancer cells or activated T CD4⁺ cells), they need not only energy, but also intense synthesis of proteins, DNA and lipid building blocks (16, 17). Although glycolysis provides less ATP compared to oxidative phosphorylation, intermediate byproducts of glycolysis are utilized as substrates for protein, DNA, and lipid biogenesis. After completing the step-by-step reasoning of similar metabolic programs in cancer and T CD4⁺ cells, could it be concluded that the same molecules control this strategy?

After activation of T CD4⁺ cells, according to variable signals of immunological microenvironment, they differentiate into Teff or Treg subsets. As shown, Teff subsets (Th1, Th2 and Th17) and Treg have different metabolisms (23-29) (Figure 1). The mechanistic target of rapamycin (mTOR) is a conserved molecule from yeast to humans. mTOR is one of the important kinases which regulate cell growth and controls cell metabolism in response to environmental signals. In mammalian cells, mTOR is encoded as a single gene. The mTOR protein consists of two complexes: mTORC1 and mTORC2 (30). These complexes play distinct roles. mTORC1 regulates autophagy, transcription, translation, and ribosome biogenesis. mTORC2 regulates cell survival. Also, cytoskeleton organization mTORC1 is the down-stream of AKt while mTORC2 is the up-stream of AKt. mTOR is a key sensor of cell energy status and nutrition. In T CD4⁺ cells, mTOR is activated by IL-2 and/or CD28 signaling (31, 32). Its activity is also controlled by metabolic signals. Consequently, both immunological and metabolic signals control mTOR activity. It is not surprising that metabolic status can affect T CD4⁺ cell function since the same molecules or pathways like mTOR are involved in immunological and metabolic signals.

For instance, CD28 signaling cascades can finally activate mTOR (17). On the other hand, an increasing level of intracellular amino acids like glutamine also activates the mTOR cascade (33).

An interesting point is that even in the presence of CD28 signaling, rapamycin-treated T CD4⁺ cells can become anergic.

Figure 1 - Metabolic differences between T CD4⁺ subsets. After activation, all differentiated T CD4⁺ subtypes change their metabolic program. This change is accompanied by an increase of metabolic demands. The important point is that all differentiated subtypes show more metabolic machinery activation compared to basic metabolism of naïve T CD4⁺. But among them, Treg cells are specific. Th1, Th2 and Th17 are initially glycolytic while Treg cells metabolism relies on lipid oxidation. Moreover, while Treg cells are less active than the others, they are more active than naïve T cells. The difference between them seems logical, because these cells get activated in different immune microenvironments and their functions are also different.
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**REVIEW**

Metabolism of cancer cells like Teff cells is glycolytic. mTOR activation results in the promotion of glycolysis in both cells. It should be noted that specific metabolites can regulate mTOR activity and consequently the fate of T CD4+ cells (Table I) (10, 15, 19, 35-46).

As shown in Table I, several studies have been devised to find the mechanisms involved in determining T CD4+ cells differentiation, and factors like fatty acids and vitamins have been evaluated. Fatty acids are one of the most important elements in the diet. The level of fatty acids intake or their ratio, for instance omega 6/omega 3, plays a critical role in body health. Palmitic, linoleic, stearic, oleic, and arachidonic acids contain 80% of phosphatidylcholine in human plasma (47, 48). The possibility exists that these fatty acids influence our immune system like other nutritional elements. Besides, previous studies were not conclusive about the role of SCFA, SFA or unsaturated fatty acid (USFA) on T CD4+ cell differentiation.

**FATTY ACIDS EFFECTS ON T CD4+ DIFFERENTIATION**

The effect of fatty acid on T CD4+ is not fully understood yet. Some studies have suggested that short chain fatty acids (SCFA), like acetate, propionate and butyrate (C2, C3 and C4 respectively) can induce effector (39) and regulatory T CD4+ cell subsets differentiation (39, 42), whereas long chain fatty acids have different effects on T CD4+ cells. Omega 3 poly unsaturated fatty acids (n-3 PUFA) are TH1 and Th17 antagonist (1) and decreased the number of cells in some osteoarthritis models (1). Furthermore, n-3 PUFA can suppress T cell function and increase Treg cells number. One possible mechanism that explains the effect of n-3 PUFA on T CD4+ is the involvement of PPAR receptors (49).

Fatty acid synthesis also plays an important role in T CD4+ differentiation. Studies have shown that inhibition of enzymes, which are engaged in fatty acid synthesis, results in Treg differentiation (50). Even at the TH17-polarizing condition, inhibition...
of fatty acid synthesis prevents TH17 differentiation (51).

It can be concluded that n-3 PUFAs have an anti-inflammatory effect (43) on the immune system. In contrast, saturated fatty acids have a pro-inflammatory impact on the immune system (52, 53). The effect of mono saturated fatty acid (MFA) on the immune system is not clearly demonstrated. Omega 6 or omega 9 MFA have different or even opposite effects on the immune system (54), a finding that may be due to the type of study design, *in vivo* or *in vitro*. Adding fatty acids to T cell or PBMC cell culture may have divergent results on T CD4+ differentiation. The level of micro and macro elements and their ratio (glucose, proteins or fatty acid) are also crucial. Results have shown that if adequate glucose is present in cell culture as an energy source, fatty acids are not consumed. Previous studies have shown that even with the presence of sufficient amounts of alternative energy sources, T CD4+ cells prefer

<table>
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<tr>
<th>Table I - Different metabolites effect on immune system.</th>
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<tr>
<td><strong>Metabolite</strong></td>
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<tr>
<td>Vitamin A</td>
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<tr>
<td>Palmitic and oleic acid</td>
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<td>Glucose (depletion)</td>
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<tr>
<td>Glucose</td>
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<td>Oxygen</td>
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<td>NaCl</td>
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<td>Palmitic acid</td>
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<tr>
<td>Acetate Propionate Butyrate (SCFA)</td>
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<tr>
<td>Omega-3 PUFA</td>
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<td>Acetate Propionate</td>
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<td>Fish oil plus astaxanthin</td>
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<td>Butyrate</td>
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HIF1: Hypoxia-inducible factor is a transcription factor, which is sensitive to decreased O2 in cell environment, NFκB: nuclear factor kappa-light-chain-enhancer of activated B cells is a controller of DNA transcription, MAPK: Mitogen-activated protein kinase is a kinase, which is involved in proliferation of cells, SCFA: short chain F.A, PUFA: poly unsaturated F.A, HDAC: Histone deacetylases, SFA: saturated F.A, MQ: macrophage and DCs: dendritic cells.
to use glucose. Although Treg cells rely on lipid oxidation, the addition of only fatty acid to the cell culture is not sufficient for Treg induction. However, the preferential energy source for T CD4+ cells is demonstrated to be glucose (55). Hence, T CD4+ subsets have various metabolic programs. Differentiated T CD4+ cells like cancer cells. Differentiated T cells have glycolytic and oxidative metabolic programs. Metabolic and immunological signals can affect the differentiation of T CD4+ cells. Metabolism plays the key roles in Th cells differentiation. Differentiation metabolic elements can potentially influence the immunometabolism. Current scientific evidence suggests that nutritional elements strongly on immunometabolism. Current research focuses on the role of nutritional elements on immunometabolism.

**CONCLUSIONS**

In the past few years, researchers focused strongly on immunometabolism. Current scientific evidence suggests that nutritional elements can potentially influence the immune system and the general body health as well. Metabolic and immunological signals can affect the differentiation of T CD4+ cells like cancer cells. Differentiated T CD4+ subsets have various metabolic programs. Among them, only Treg depends on lipid oxidation for differentiation. It is important to note that for *in vitro* Treg induction alongside the addition of fatty acid, glucose deprivation is necessary.

**REFERENCES**