Seminars in Immunology 28 (2016) 425–430

Contents lists available at ScienceDirect

Seminars in Immunology

journal homepage: www.elsevier.com/locate/ysmim

Review

Immunometabolic circuits in trained immunity

Rob J.W. Arts, Leo A.B. Joosten, Mihai G. Netea*

Department of Internal Medicine and Radboud Center for Infectious Diseases (RCI), Radboud University Medical Center, Nijmegen, The Netherlands

ARTICLE INFO

Article history:
Received 12 July 2016
Received in revised form
16 September 2016
Accepted 19 September 2016
Available online 27 September 2016

Keywords:
Monocyte
Macrophage
Glycolysis
Epigenetics
Trained immunity
Immunometabolism

ABSTRACT

The classical view that only adaptive immunity can build immunological memory has recently been challenged. Both in organisms lacking adaptive immunity as well as in mammals, the innate immune system can adapt to mount an increased resistance to reinfection, a *de facto* innate immune memory termed *trained immunity*. Recent studies have revealed that rewiring of cellular metabolism induced by different immunological signals is a crucial step for determining the epigenetic changes underlying trained immunity. Processes such as a shift of glucose metabolism from oxidative phosphorylation to aerobic glycolysis, increased glutamine metabolism and cholesterol synthesis, play a crucial role in these processes. The discovery of trained immunity opens the door for the design of novel generations of vaccines, for new therapeutic strategies for the treatment of immune deficiency states, and for modulation of exaggerated inflammation in autoinflammatory diseases.

Contents

1. Introduction

Classically, the immune system can be divided into innate and adaptive immunity, with monocytes, macrophages, neutrophils, and NK cells as the main cellular effectors of innate immune responses, while T- and B-lymphocytes mediate adaptive immunity. Until recently it was assumed only the adaptive immune system possesses the capacity to mount a memory response and therefore improve the immunological reaction to a second infection. However, an increasing amount of evidence accumulates, suggesting that also the innate immune system possesses adaptive characteristics. In plants and non-vertebrates, both lacking an

Abbreviations: Ac, acetylation; ATP, adenosine triphosphate; BCG, Bacillus Calmette Guerin; CIC, citrate carrier; GABA, γ-aminobutyric acid; GAPDH, glyceraldehyde 3-phosphate dehydrogenase; H3K, histone 3 lysine; Hif, hypoxia inducible factor; HDAC, histone deacetylase; LDH, lactate dehydrogenase; me3, trimethylation; NAD(P), nicotinamide adenine dinucleotide (phosphate); OxPhos, oxidative phosphorylation; PDH, pyruvate dehydrogenase; PKM, pyruvate kinase; PPP, pentose phosphate pathway; TCA, tricarboxylic acid cycle.

* Corresponding author at: Department of Internal Medicine (463), Radboud University Nijmegen Medical Centre, Geert Grooteplein Zuid 8, 6525 GA, Nijmegen, The Netherlands.

E-mail addresses: rob.jw.arts@radboudumc.nl (R.J.W. Arts), leo.joosten@radboudumc.nl (L.A.B. Joosten), mihai.netea@radboudumc.nl (M.G. Netea).

http://dx.doi.org/10.1016/j.smim.2016.09.002

adaptive immune system, it has already been know for several decades that a memory response could be build that would protect from a secondary infection [1,2]. Interestingly, this improved secondary response is not always strictly specific, as infection with one pathogen can sometimes also protect from infections with non-related pathogens [3]. This process has been shown to be epigenetically regulated [4]. In retrospect, this (non-specific) memory, mediated by the innate immune system, was also seen in mice strains with known deficiencies in adaptive immune responses [3,5].

More recently, this same process of innate immune memory was also been shown to occur in human monocytes and macrophages. β-glucan, a major component of the *C. albicans* cell wall, was shown to *ex vivo* enhance cytokine production to a second unrelated stimulus, a process that was also epigenetically regulated [6,7]. Also in mice, an *in vivo* challenge with β-glucan protected from subsequent *C. albicans* or *S. aureus* infection [6,8], and infection with cytomegalovirus induced improved effector function of NK cells after the infection [9]. In humans, vaccination with Bacillus Calmette-Guérin (BCG) showed non-specific protection from all-cause mortality, mainly a result of reduced mortality from infections, in low-weight birth children in West-Africa [10,11]. In a vaccination study in healthy adult volunteers, BCG markedly increased *ex vivo* cytokine responses by inducing epigenetic reprogramming in myeloid cells [12]. These studies demonstrate that the innate immune system can adapt after a previous challenge through functional and epigenetic reprogramming, a process that has been termed *trained immunity* or *innate immune memory* [13,14]. On the one hand, trained immunity is likely an important host defense mechanism contributing to the maturation of the innate immune system of infants and mediating protection after certain infections or vaccinations. On the other hand, when induced inappropriately by endogenous stimuli, trained immunity may play a role in the pathogenesis of autoinflammatory and/or autoimmune diseases [14,15]. Therefore, better understanding of the molecular mechanisms of trained immunity is crucial for developing new ways of immunotherapy and targeting inflammatory disorders [14].

In the last years an increasing amount of evidence has been accumulated in support of the concept that cellular metabolism is correlated with the functional state of immune cells [16]. Some of the first observations reported that different subsets of lymphocytes had distinctly different cellular metabolic states. Activated T lymphocytes have high rates of both glycolysis and oxidative phosphorylation (OxPhos) and metabolize glucose to lactate [17,18], whereas memory T-lymphocytes are more dependent on lipid synthesis via mitochondrial citrate production. These lipids can be used to produce triacyglycerides (TAGs) which are being degraded by β-oxidation to fuel OxPhos via acetyl-CoA production [19]. In contrast, regulatory T-cells fuel β-oxidation and OxPhos through exogenously derived fatty acids [20]. This shows that the phenotype of lymphocytes highly correlates with the source of energy that they use [21,22]. Subsequent studies showed that different phenotypes of macrophages retrieve energy from very distinct metabolic pathways: the more inflammatory (M(IFNγ) or formerly M1) macrophages depend largely on glycolysis and show impaired OxPhos and disruption of the Tricarboxylic acid (TCA) cycle [23,24], while in the more anti-inflammatory (M(IL-4) or formerly M2) macrophages the Krebs cycle is intact as they rely on OxPhos [25,26] and furthermore show increased β-oxidation as a result of fatty acid uptake [27].

In this review we will focus on the metabolic pathways induced by innate immune training. We will discuss glycolysis, TCA cycle, glutamine, and cholesterol metabolism and discuss their (potential) effect on epigenetics and how these clues could be used as therapeutic targets.

2. Metabolic pathways in trained immunity

2.1. Glycolysis

A metabolic switch from oxidative phosphorylation to glycolysis resulting in more lactate production has been described in the highly metabolically active cancer cells already in the beginning of the last century by Otto Warburg, called thus the 'Warburg effect' [28]. Glycolysis is often increased during immune cell activation: activated T cells show increased rates of glycolysis [17,18] and proinflammatory macrophages increase glucose metabolism resulting in increased lactate production [24]. This switch is assumed to be important as glycolysis, although being less efficient in generating adenosine triphosphate (ATP), can be upregulated multiple folds and therefore results in a faster production of ATP compared to oxidative phosphorylation [28].

A similar switch is seen in β-glucan trained monocytes [29] (Fig. 1). Transcriptional and epigenetic (H3K4me3 and H3K27ac) analysis of β-glucan induced trained immunity in monocytes revealed that genes in the mTOR signalling pathway and several metabolic pathways, especially glycolysis, were highly induced [7,29]. When human monocytes are stimulated *in vitro* with β-glucan for 24 h and let to rest for 6 subsequent days, the amounts of glucose consumption and lactate production increased over time. In contrast, oxygen consumption 6 days after β-glucan training is significantly decreased compared to control macrophages [29]. These effects of β-glucan-induced training were mediated by activation of the Akt/mTOR/Hif1α pathway. Inhibiting this pathway at several levels, or making use of myeloid cell specific Hif1α knockout mice, abrogated induction of trained immunity both at cytokine and epigenetic level [29] (Fig. 1).

Induction of glycolysis results in higher ratios of NAD^+/NADH ratio, which was also the case in β-glucan trained monocytes/macrophages [29]. In LPS stimulated monocytes the vast increase in NAD^+/NADH ratio has been shown to activate sirtuin 1 and 6, supporting a switch from a proinflammatory state with high rates of glycolysis to a more anti-inflammatory state with increased fatty acid oxidation [30]. Interestingly, in β-glucan trained monocytes, sirtuin 1 expression appeared to be decreased and activation of sirtuin 1 by resveratrol inhibited training [29]. This suggests that decreased expression of sirtuin 1 might play a role in the significant increase of H3K27ac induced by monocyte training by β-glucan. However, apart from the classical role as histone deacetylases, sirtuins were also shown to deacetylate nonhistone structutes, such as NF-κB or Hif1α [31], and this should be taken into consideration too. Moreover, lactate, the end product of anaerobic glycolysis, is also able to inhibit histone deacetylase (HDAC) activity and therefore cause increased gene accessibility [32].

In addition to its direct role in energy production, induction of glycolysis might also play a role in posttranslational modification of effector molecules. Glyceraldehyde 3-phosphate dehydrogenase (GAPDH), one of the enzymes in glycolysis can bind the 3′ UTR of *ifng* RNA and therefore decreasing IFNγ production. In activated T cells GAPDH is used in glycolysis and therefore more IFNγ can be produced. Also decreasing GAPDH expression by RNA interference increased IFNγ production [33]. A similar mechanism has been reported in murine and human monocytes and macrophages in relation to TNF mRNA induction. GADPH can posttranscriptionally repress TNF mRNA in monocytes in low glycolysis state, e.g. immunotolerant monocytes as seen in sepsis [34] (Fig. 2). Whether reversal of such effects plays a role in trained immunity remains to be elucidated.