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Regulation of T Cell Immunity by Dendritic Cells

Minireview

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Immune responses are initiated in the T cell areas of secondary lymphoid organs, where naïve T lymphocytes encounter dendritic cells (DCs) that present antigens taken up in peripheral tissues or locally. Antigens that do not have access to DCs are thus ignored by T cells, while those that do have access to DCs can stimulate naïve T cells, driving their proliferation and differentiation or, in special circumstances, their deletion. Thus, DCs represent the interface between the universe of foreign and tissue-specific antigens and T lymphocytes and are the key players in the regulation of cell-mediated immunity.

In this review, we will discuss how DCs provide a quantitative and qualitative framework for T cell antigen recognition. We will first summarize the requirements for activation and differentiation of naïve T cells in terms of peptide-MHC complexes, costimulatory molecules, and cytokines. Then, we will consider how DCs assemble these components into discrete packets that are delivered to the T cell areas. Finally, we will discuss how the density and quality of antigen-carrying DCs might determine the magnitude and class of T cell responses.

Requirements for Activation and Differentiation of Naïve T Lymphocytes

To recognize antigen, T cells need to establish contact with antigen-presenting cells (APCs) by forming an immunological synapse, where T cell receptors (TCRs) and costimulatory molecules are congregated in a central area surrounded by a ring of adhesion molecules (Dustin and Cooper, 2000). At the synapse, a serial TCR triggering by peptide-MHC complexes initiates a signaling cascade of which the magnitude and duration determine the entry of naïve T cells into the cell cycle. Synapses form within minutes after TCR triggering and are stable in the absence of disturbing events, but can be disrupted by cell division, death of APCs, or by external influences, such as collagen. It is important to consider that T cells are continuously poised to form better synapses and can rapidly shift from one APC to another, offering a higher level of stimulation.

The amount of signal that T cells receive is dependent on three factors: (1) the level of peptide-MHC complexes that initiate signal transduction, (2) the level of costimulatory molecules that amplify the signaling process, and (3) the stability of the synapse that determines for how long the signaling process is sustained (Lanzavecchia and Sallusto, 2001). The efficiency of the synapse as a signal transducing machinery varies with the nature of the APC and the T cell's developmental stage. In activated, effector, and memory T cells, TCR triggering is efficiently coupled to signal transduction pathways so that the cells can rapidly respond to low doses of antigens even in the absence of costimulation. In contrast, in naïve T cells, TCRs are inefficiently coupled to downstream signal transduction pathways. Engagement of CD28 by B7 molecules expressed on APCs recruits membrane rafts containing kinases and adapters to the synapse and amplifies up to 100-fold the signaling process initiated by the TCRs. Thus, in the absence of costimulation, naive T cells can be activated only by extremely high (nonphysiologic) doses of antigens and require a prolonged stimulation. In contrast, in the presence of costimulation, they can respond to ~100-fold lower doses of antigen and also more rapidly. Depending on the antigen dose and level of costimulation, naïve T cells require 6 to greater than 30 hr of TCR stimulation to reach commitment, while memory/effector T cells respond within 0.5 to 2 hr.

Once committed to the first division, T cells proliferate rapidly in response to IL-2, which is produced by activated T cells and can act in autocrine and paracrine fashions. Since IL-2 production is induced by antigenic stimulation and is greatly enhanced by costimulation, its availability in T cell areas varies widely as a function of the extent and duration of T cell activation.

There is growing evidence that the duration of TCR stimulation, together with polarizing cytokines, determines the progressive differentiation of $CD4^+$ T cells, leading to the generation of terminally differentiated effector cells as well as intermediates (Lanzavecchia and Sallusto, 2000). $CD4^+$ T cells that receive a short TCR stimulation in the absence of IL-12 proliferate but do not differentiate to effector cells. Upon in vivo transfer, they home to the lymph nodes and, following secondary antigenic challenge, differentiate to effector cells. In contrast, T cells that receive a prolonged TCR stimulation in the presence of IL-12 or IL-4 terminally differentiate to Th1 cells, producing IFN-γ, or to Th2 cells, producing IL-4, IL-5, and IL-13. As part of their differentiation program, Th1 and Th2 cells lose the lymph node homing receptors and acquire the capacity to migrate to inflamed nonlymphoid tissues to execute their effector functions. The progressive differentiation model is supported by the existence of a distinct population of central memory T cells that lack immediate effector function and carry the lymph node homing receptor CCR7.

Recent experiments indicate that, once commitment has been reached, $CD8^+$ T cells can autonomously divide (at least 7 times) and acquire cytotoxic function in the absence of further antigenic stimulation (Kaech and Ahmed, 2001; van Stipdonk et al., 2001). These findings imply that the clonal expansion and differentiation programs are imprinted in naïve $CD8^+$ T cells during a short encounter with APCs, a fact that may reflect a higher propensity to undergo terminal differentiation. It is unclear, however, which stimuli may control the generation of different types of effector and memory $CD8^+$ T cells as well as their peripheral deletion.

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DC Maturation: Assembling Packets of Information for T Cells

DCs that migrate from tissues to lymph nodes have a life expectancy of 2–3 days and can therefore be viewed as disposable packets, each carrying a given amount of peptide-MHC complexes, costimulatory molecules, and cytokines. These packets are assembled during DC maturation, a process which is initiated by pathogens and/or inflammatory stimuli. Maturation coordinately regulates DCs' antigen capturing, processing and presentation, expression of costimulatory molecules, cytokine production, and life-span.

The variety and number of antigenic determinants presented by an APC is dependent on the efficiency of antigen capture and on the availability of MHC molecules to be loaded with antigenic peptides. Immature DCs possess high levels of constitutive macropinocytosis and express endocytic receptors for pathogens, such as the mannose receptors, DEC-205 and DC-SIGN. Maturation transiently increases synthesis and transport of MHC class II molecules, while decreasing their degradation, thus favoring the rapid accumulation of many peptide-MHC complexes, which are retained for several days while class II synthesis is shut off. Presentation on MHC class I molecules is also enhanced due to an $\sim$10-fold increase in the rate of MHC class I synthesis, which is sustained in mature DCs. DCs are also capable of transporting antigens from the endocytic compartment to the cytosol, leading to "cross-presentation" on MHC class I molecules to $CD8^+$ T cells. Thus, by coupling antigen capturing and processing to MHC class II synthesis and transport, maturing DCs assemble large numbers of MHC molecules loaded with antigenic peptides, thus maximizing TCR stimulation.

Maturation stimuli upregulate the costimulatory molecules B7.1 and B7.2, which are transported together with MHC class II molecules to the cell surface where they remain associated within membrane microdomains (Turley et al., 2000), a condition that may enhance the effectiveness of TCR and CD28 triggering. The simultaneous increase of MHC and B7 molecules synergistically enhances the T cell stimulatory capacity of DCs. On the one hand, the upregulation of MHC molecules ensures high capacity for antigen presentation, i.e., presentation of more epitopes, each at a high copy number. On the other, B7 upregulation ensures an effective amplification of signaling in naïve T cells.

Cytokine production by DCs is subject to a tight regulation, which is particularly relevant in the case of IL-12, the prototypic Th1-polarizing cytokine. IL-12 production is elicited by most pathogens and is potently boosted by activated T cells through CD40L (Schulz et al., 2000). However, IL-12 is not induced by some maturation stimuli, such as TNF-α, IL-1, cholera toxin, or FasL. IL-12 production can be modulated by cytokines and mediators present during induction of maturation (Kalinski et al., 1999). Thus, IFN-γ and even IL-4 enhance IL-12 production induced by appropriate stimuli, while prostaglandin E2 and IL-10 exert an inhibitory effect. Moreover, IL-12 production by DCs is restricted to a narrow temporal window (8–16 hr) after induction of maturation (Langenkamp et al., 2000). Thus, the Th1-polarizing capacity of DCs is contingent on a number of variables that include the microenvironment, the maturation stimuli, and the kinetics of maturation. In conclusion, the DC maturation process results in the production of specific packets that contain, in variable combinations, the essential elements required for T cell activation and polarization.

Dynamic Changes in DC Populations Impacting on T Cell Responses

Because the half-life of mature DCs is short and because cytokine production is transient, the number and properties of DCs present in the T cell areas reflect, in a sensitive and dynamic manner, the conditions of the tissues from which the lymph is drained. We discuss below two possible scenarios.

Under steady-state conditions, a small fraction of resident DCs "spontaneously" mature and migrate to the draining lymph nodes, carrying antigens and apoptotic bodies taken up in peripheral tissues (Huang et al., 2000). These DCs can either present antigen directly or release it so that it is presented, at lower levels, by other DCs (Inaba et al., 1998). There is increasing evidence that this presentation pathway leads to T cell deletion and to peripheral tolerance (Heath and Carbone, 2001). Indeed, under steady-state conditions, $CD8^+$ T cells specific for a tissue antigen are activated by migratory APCs, most likely DCs, in the draining lymph nodes, but undergo only a few divisions and die without acquiring effector function. There may be several mechanisms that contribute to this effect. Spontaneously matured DCs may deliver to T cells a qualitatively distinct tolerizing signal. Alternatively, the low frequency and short lifespan of DCs, together with the low level of antigen and B7, may deliver a weak and transient stimulation, which is not sufficient to initiate T cell proliferation and differentiation. Moreover, the low levels of IL-2 available under these conditions may fail to sustain proliferation of primed T cells. Immune responses to poorly immunogenic antigens can be enhanced by antigen-specific $CD4^+$ T cells and even by antibodies to CD40 that act by increasing the T cell stimulatory capacity of antigen-carrying DCs. Lymph node homing central memory T cells are particularly effective in DC activation and may jump-start secondary responses, even if DCs are poorly stimulatory.

When pathogens (or adjuvants) are present in peripheral tissues, resident DCs are activated en masse and migrate to the draining lymph nodes. At the same time, monocytes are recruited from peripheral blood into inflamed tissues. There, they rapidly differentiate to antigen-capturing DCs and, upon maturation, migrate to the draining lymph nodes, thus sustaining antigen sampling and presentation for extended periods of time. Maturing DCs produce large amounts of inflammatory cytokines and chemokines that promote and sustain monocyte recruitment and, at the same time, upregulate CCR7, which is required for their migration into lymphatic vessels and their localization to the T cell areas. The relative role of tissue-resident DCs, such as Langerhans cells and dermal DCs, versus recruited DCs, such as monocyte-derived DCs and, possibly, plasmacytoid DCs (pDCs), remains to be established. Production of IFN I by pDCs may be important to promote maturation of myeloid DCs (mDCs) and protect them from the cytopathic effects of viruses (Cella et al., 2000; Kadowaki et al., 2000).

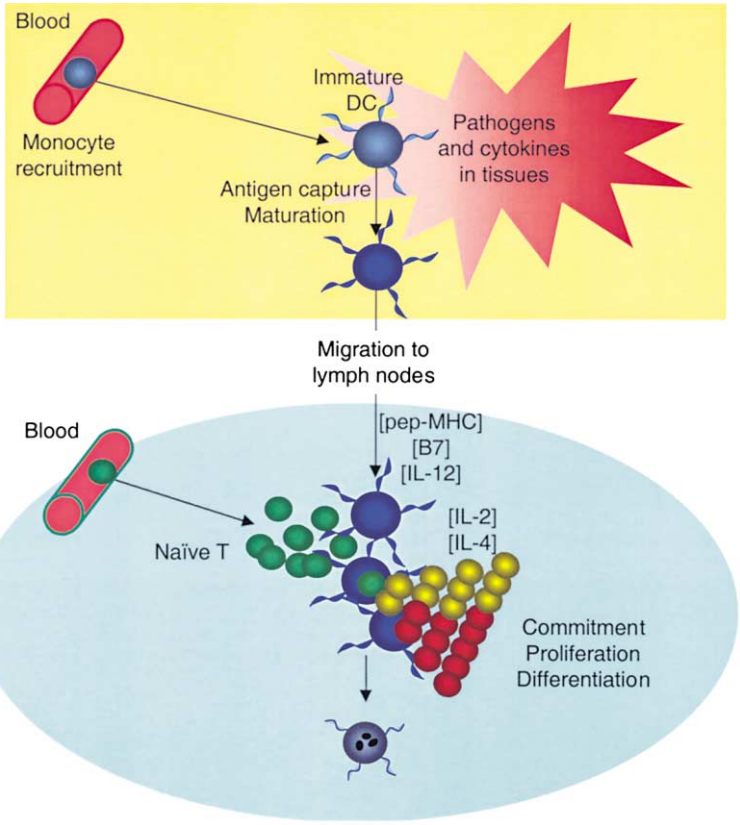

Figure 1. T Cell Priming by DCs

Recruitment of DC precursors (monocytes) into peripheral tissues and maturation of DCs in response to pathogens or cytokines result in migration to the draining lymph node of large numbers of DCs carrying high levels of peptide-MHC complexes and B7. By physical interaction through an immunological synapse, naïve T cells (green) achieve stimulation and become committed to proliferate in response to IL-2. Sustained TCR stimulation by continuous contact with DCs and polarizing cytokines (IL-12 and IL-4) promote T cell differentiation to nonlymphoid tissue-homing effector cells (red). T cells receiving a shorter stimulation do not acquire effector function and retain lymph node-homing capacity (yellow). Mature DCs have a short life-span and die within 2–3 days. As a consequence, the composition of DCs in the lymph nodes dynamically reflects what is occurring in the peripheral tissues.

In summary, under inflammatory conditions, the T cell areas receive large numbers of highly stimulatory DCs for a sustained period of time (Figure 1). The high DC density and the high levels of antigen and B7 molecules deliver a strong and sustained stimulation to specific T cells, leading to their rapid commitment to proliferation and differentiation. Under these conditions, high levels of IL-2 are produced, which drive clonal expansion of committed T cells irrespective of whether or not they continue to receive TCR stimulation. DC-T cell interaction results in a reciprocal stimulation. Activated T cells trigger DCs via CD40L or TRANCE, improving their T cell stimulatory capacity, boosting IL-12 production and prolonging their lifespan. In contrast, anergic or regulatory T cells may suppress antigen presentation by DCs via production of inhibitory cytokines or direct contact.

A long-standing question is whether different subsets, such as murine mDCs and lymphoid DCs, are endowed with unique capacities to induce Th1 or Th2 responses (Moser and Murphy, 2000). As discussed above, mDCs produce IL-12 only in response to some pathogens and CD40L, and within a narrow time window. In addition pDCs produce large amounts of IFN I, another Th1 polarizing cytokine, in response to viruses but not to CD40L and, again, only within a narrow time window. Thus, the Th1 polarizing capacity of both mDCs and pDCs is contingent upon appropriate stimulation and timing. In contrast, the capacity to induce Th2 responses is a property of DCs that do not produce Th1-polarizing cytokines, either because they have been conditioned by nonpermissive stimuli or because they have exhausted their IL-12- or IFN I-producing capacity under these conditions. Th2 polarization can be driven by IL-4, which is produced by T cells themselves or may be derived from exogenous sources, such as NK-T cells. It has been suggested that the dynamics of DC migration to the draining lymph nodes may result in rapid changes in DC composition and cytokine content in T cell areas. This may lead to preferential priming of Th1 cells during the early phases of the immune response, when recently stimulated DCs enter the T cell areas in large numbers. This may be followed by priming of Th2 and nonpolarized T cells at later time points, when the influx of DCs ceases and the DCs surviving in the T cell area exhaust their IL-12 producing capacity (Lanzavecchia and Sallusto, 2000).

There are other aspects of the immune response in which DCs may play an important role. DCs are present in germinal centers, suggesting their participation in B cell stimulation. Signals from DCs via OX40L upregulate, in responding T cells, CXCR5, a receptor which drives helper T cells to the B cell follicles (Lane, 2000). Furthermore, IFN I produced by pDCs can potently enhance antibody responses and isotype switching by stimulating DCs in vivo (Le Bon et al., 2001). Finally, recent studies indicate that immature DCs prime regulatory T cells producing IL-10 (Dhodapkar et al., 2001; Jonuleit et al., 2000), while pDCs prime T cells for production of IL-10 and IFN-γ (Cella et al., 2000; Kadowaki et al., 2000).

The availability of antigen-presenting DCs and of antigen-specific T cell precursors represents the limiting factors in the immune responses. There is growing evidence that responding T cells compete in vivo for access to DCs and that this competition can be relieved by

providing more DCs (Kedl et al., 2000). At the initial phase of a primary response, the low frequency of naïve T cells specific for a given antigen makes competition among responding cells unlikely. However, as the responding cells proliferate, competition for sustained TCR stimulation will increase, particularly among cells of the same clone, which have the same avidity and occupy the same niche. This intraclonal competition may drive functional diversification: T cells achieving a sustained stimulation differentiate to effector cells, while those receiving a short stimulation remain in an intermediate state giving rise to central memory T cells. In contrast, in secondary responses, specific T cells are present at high frequency and may directly compete for access to antigen-carrying DCs. This process of interclonal competition may explain the selection of high avidity T cells which occurs during secondary responses.

In conclusion, DCs provide the adaptive immune system with the essential function of context discrimination. Within individual DCs, multiple stimuli from pathogens and inflammatory cytokines are integrated into distinct outputs in terms of antigen presentation, costimulation, and cytokine production. The same stimuli also recruit large numbers of DC precursors, thus leading to sustained antigen sampling in peripheral tissue and presentation to T cells in lymph nodes. The T cell activation and differentiation program is tailored to discriminate antigen concentration, cytokine and costimulatory molecule composition, and DC density, leading to the generation of appropriate T cell responses ranging from tolerance to inflammation, cytotoxicity, and memory.

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