Immunity
Review

In Vivo Induction of Immune Responses to Pathogens by Conventional Dendritic Cells

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Specific defense mechanisms against pathogens are fulfilled by different subsets of nonmucosal conventional dendritic cells (DCs), including migratory Langerhans cells (LCs), dermal DCs, and resident $CD8^+$ and $CD8^-$ DCs found in lymphoid organs. Dermal DCs capture antigens in the skin and migrate to lymph nodes, where they can transfer the antigens to $CD8^+$ DCs and activate $CD4^+$ T cells. Differential antigen-processing machinery grants $CD8^+$ DCs a high efficiency in activating $CD8^+$ T cells through crosspresentation, whereas $CD8^-$ DCs preferentially trigger $CD4^+$ T cell responses. Recent findings have revealed the important role played by monocyte-derived DCs (mo-DCs), newly formed during infection, in activating $CD4^+$ and $CD8^+$ T cells, regulating immunoglobulin production, and killing pathogens. However, a number of controversial issues regarding the function of different DC subsets during viral, bacterial, and parasitic infections remain to be resolved.

Introduction

Over the last decade, mouse experimental models of infection have provided important insights into the identification, origin, and functional specialization of different dendritic cell (DC) subsets (reviewed by Shortman and Naik, 2007). Specific functions related to defense mechanisms against pathogens have been attributed to the main subsets of mouse conventional DCs (cDCs), which include migratory epidermal Langerhans cells (LCs) and dermal DCs, as well as resident $CD8^+$ and $CD8^-$ DCs found in peripheral lymph nodes (per-LNs) and the spleen (reviewed by Villadangos and Schnorrer, 2007). However, these reports have also led to controversies over the functional specialization of different DC subsets, probably because of the fact that currently available information relies on studies that explored DC-mediated immune responses against a limited number of pathogens and that were performed with widely diverse experimental strategies. In addition, the recent finding that inflammatory monocyte-derived DCs (mo-DCs) are formed de novo during infectious processes at infection sites and lymphoid organs (reviewed by Leon and Ardavin, 2008b) adds an extra level of complexity to the definition of specific functional capacities of DC subsets. Here, we will discuss the current understanding of the functional relevance of nonmucosal cDCs during in vivo immune responses against viruses, bacteria, and parasites.

Migratory and Resident cDCs in Steady State

cDCs involved in defense mechanisms against pathogens comprise all the DC subsets located in antigen-capture areas related to epithelial surfaces (such as the skin and the mucosa of the digestive, respiratory, and reproductive tracts) and secondary lymphoid organs (such as the spleen, LNs, and gut- and bronchial-associated lymphoid tissues). Because the role of mucosal cDCs has been covered in recent reviews on intestinal, pulmonary, and vaginal cDCs (de Heer et al., 2005; Iwasaki, 2007; Johansson and Kelsall, 2005), we will focus on the role of nonmucosal cDCs in the induction of in vivo immune responses against pathogens. Nonmucosal cDCs include those located in the skin, per-LNs, and spleen, and they can be subdivided into two categories: migratory cDCs and resident cDCs (reviewed by Shortman and Naik, 2007).

Skin migratory cDCs participate in defense mechanisms against pathogens that gain access to the epidermis or the dermis as a consequence of pathogen-specific skin-invasive mechanisms, skin lesions, or inoculation by insects, such as the malaria mosquito or *Leishmania* sandfly. In steady state, migratory cDCs include immature epidermal LCs and immature dermal DCs that remain in the skin until they migrate, in the absence of inflammation or infection, into the per-LNs through the lymph. This migration process involves the upregulation of MHC class II and costimulatory molecules on the surface of LCs and dermal DCs; this implies that in steady state within the per-LNs, these two DC subsets appear in a mature state with regards to their expression of MHC class II and costimulatory molecules (reviewed by Villadangos and Schnorrer, 2007). LCs have been demonstrated to derive from monocytes (Ginhoux et al., 2006). Although indirect evidence also supports a monocytic origin for dermal DCs (reviewed by Leon and Ardavin, 2008b), this hypothesis remains to be conclusively demonstrated. Within the per-LNs of C57BL/6 mice, LCs and dermal DCs represent approximately 35% and 25% of total cDCs, respectively (Kamath et al., 2002). Analyses of BrdU-labeling kinetics of LCs and dermal DCs within the per-LNs of C57BL/6 mice (Kamath et al., 2002) revealed that migratory cDCs have a slow turnover (half-life > 21 days and 12 days for LCs and dermal DCs, respectively).

Studies based on experimental models of cutaneous infection, which use non-genetically modified mice, as well as studies based on models that use genetically engineered mice and that allow LC in vivo tracking or LC conditional ablation (reviewed by Kissenpfennig and Malissen, 2006) concur on the concept that LCs are not directly involved in the induction of pathogen-specific T cell responses (Allan et al., 2003; Filippi et al., 2003; Lemos et al., 2004; Zhao et al., 2003), although a role for LCs in antigen transport to the per-LNs cannot be excluded. Nevertheless, the participation of LCs in immunity against pathogens

under physiological conditions is not fully resolved (Kissenpfennig and Malissen, 2006; Villadangos and Heath, 2005), and here, we will focus on the role of the better understood dermal DCs in immunity against pathogens.

Immature $CD8^+$ DCs and $CD8^-$ DCs are found in per-LNs and the spleen. $CD8^+$ DCs are mainly located within the T cell areas (i.e., the per-LN paracortex and spleen inner white pulp), whereas $CD8^-$ DCs are preferentially located in antigen-capture areas (i.e., the per-LN subsinusal layer beneath the subcapsular sinus, as well as the spleen marginal zone). Within the spleen of C57BL/6 mice, $CD8^+$ DCs and $CD8^-$ DCs represent approximately 25% and 75% of total cDCs, respectively, whereas in the per-LNs, each subset represents approximately 20% of total cDCs (Kamath et al., 2002). Among spleen resident cDCs, $CD8^+$ DCs have the fastest turnover (half-life: 1.5 days), indicating that in the steady state, they are replaced within 3 days, whereas $CD8^-$ DCs have a lower turnover (half-life: 3 days). In contrast, within the per-LNs, both $CD8^+$ DCs and $CD8^-$ DCs display a longer lifespan than they do in the spleen, with a half-life of 4.5 days for both subsets (Kamath et al., 2002). $CD8^-$ DCs can be further subdivided into $CD8^-CD4^-$ and $CD8^-CD4^+$ DC subsets (reviewed by Villadangos and Schnorrer, 2007), but because no exclusive functions have been specifically ascribed to these subsets regarding their role in immune responses against pathogens, we will not refer to the subsets individually.

On the basis of current knowledge, $CD8^+$ and $CD8^-$ DCs appear to fulfill their specific defense functions against pathogens essentially within the environment of the per-LNs or spleen and therefore do not appear to migrate through the blood or lymph to other organs. However, DC-activating stimuli, such as Toll-like receptor (TLR) ligands and inflammatory cytokines and chemokines, induce the maturation of resident cDCs, a process that can determine internal migratory processes from antigen-capture areas to the T cell areas within the LNs or the spleen. Interestingly, chemokine-induced migration of DCs from the splenic marginal zone to the inner white pulp was required for bacterial transport to the T cell areas and initiation of $CD8^+$ T cell responses against *Listeria monocytogenes* (Aoshi et al., 2008 [in this issue of *Immunity*]). After activation, $CD8^-$ DCs migrate from the splenic marginal zone to the inner white pulp, where they interact with antigen-specific T cells (De Smedt et al., 1996). In addition, analysis of the immune response against *Toxoplasma gondii* (Reis e Sousa et al., 1997) suggests that during infection-induced inflammatory reactions, $CD8^+$ DCs migrate from the outer to the inner splenic white pulp. This finding contrasts with the concept that in steady state, $CD8^+$ DCs are found in the inner white pulp T cell areas. It could therefore be hypothesized that during infection, newly formed $CD8^+$ DCs are first located in the outer white pulp, close to the marginal zone, and subsequently migrate to the T cell areas after contact with a pathogen. In this regard, $CD8^+$ DCs located in the T cell areas in steady state could be mainly involved in T cell tolerance induction (reviewed by Steinman et al., 2003). Both $CD8^+$ DCs and $CD8^-$ DCs appear to derive, in the steady state, from a common nonmonocytic-precursor population (Naik et al., 2006), although the differentiation of each subset is differentially regulated by specific signaling pathways (reviewed by Shortman and Naik, 2007). Resident cDCs located in the per-LNs or spleen are involved in immune responses against pathogens that gain access to these organs through the lymph or blood or against pathogens transported to the per-LNs by skin migratory cDCs. The different migratory and resident DC subsets found in steady state in the skin, per-LNs, and spleen are summarized in Figures 1 and 2. The phenotypic characteristics of these DC subsets are summarized in Table 1.

De Novo Formation of Inflammatory DCs during Infection

Studies performed during acute or chronic infections caused by viruses, bacteria, and parasites have demonstrated a dramatic increase in the number of cDCs present at both the infection sites, such as the skin, and associated lymphoid organs, such as the draining LNs or the spleen (Leon and Ardavin, 2008a; Martin et al., 2002; Serbina et al., 2003; Yoneyama et al., 2001). In these locations, inflammatory cDCs are mo-DCs derived from $Ly\text{-}6C^+$ monocytes, which are not found in the steady state (reviewed by Leon and Ardavin, 2008b). Monocytes recruited to the dermis differentiate into dermal mo-DCs, which are first located in the dermis but subsequently migrate into the per-LNs through the lymph. Inflammatory $Ly\text{-}6C^+$ monocytes are recruited to the skin through inflamed dermal venules by a mechanism involving the interaction of P-selectin glycoprotein ligand-1 (PSGL-1) with endothelial P- and E-selectins and the interaction of L-selectin with endothelial peripheral lymph node addressin (PNAd; Leon and Ardavin, 2008a). Interestingly, inflammatory dermal mo-DCs and dermal DCs found in steady state display similar phenotypic features (Leon et al., 2007), suggesting a monocytic origin for dermal DCs, as discussed above. $Ly\text{-}6C^+$ monocytes recruited to the per-LNs and spleen during infection-induced inflammation differentiate into LN mo-DCs and spleen mo-DCs, respectively. $Ly\text{-}6C^+$ monocyte migration to the per-LNs occurs through high endothelial venules by a mechanism essentially involving L-selectin and PNAd interactions (Leon and Ardavin, 2008a). To our knowledge, no data on the molecules involved in monocyte-endothelial interactions during monocyte migration through the splenic marginal sinus have been reported.

Importantly, although inflammatory DCs are considered to correspond essentially to DCs derived from $Ly\text{-}6C^+$ monocytes, a strong increase in $CD8^+$ and $CD8^-$ DC subsets has also been reported during infection (Leon and Ardavin, 2008a; Martin et al., 2002). These observations indicate that inflammatory cDCs also comprise $CD8^+$ and $CD8^-$ DCs phenotypically similar to those found in steady state, and they suggest that inflammatory $CD8^+$ and $CD8^-$ DCs are formed de novo in inflammatory foci and lymphoid organs during an infection. Several reports support the hypothesis that $CD8^+$ and $CD8^-$ DCs formed de novo under inflammation derive from $Ly\text{-}6C^+$ monocytes (Leon and Ardavin, 2008b; Leon et al., 2007; Leon et al., 2004), although this hypothesis is still a matter of controversy and has yet to be addressed conclusively. The DC subsets found under inflammatory conditions in the skin, per-LNs, and spleen are illustrated in Figures 1 and 2.

Functional Relevance of Pre-existing cDCs versus De Novo-Formed Inflammatory cDCs

Taking into account the fast turnover of per-LN and spleen cDCs (Kamath et al., 2002), the fast increase in cDC numbers observed

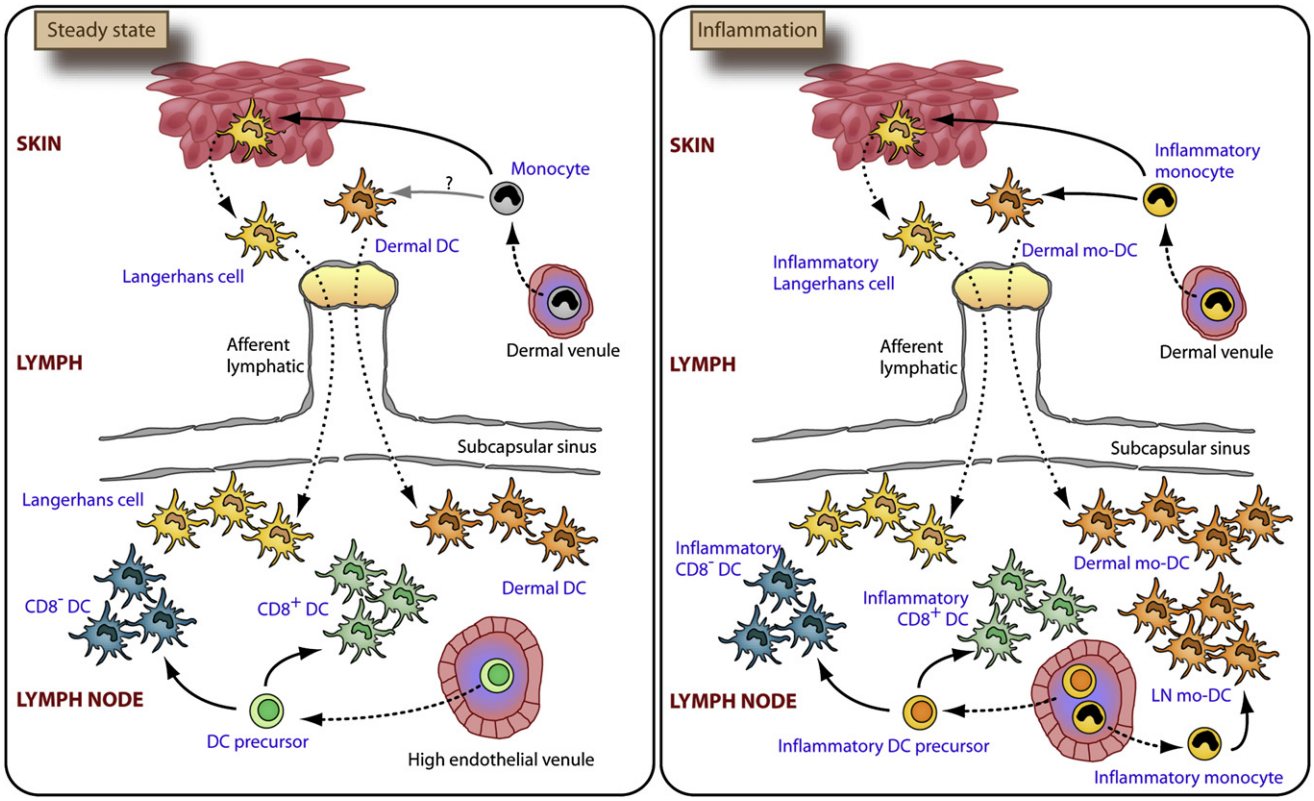

Figure 1. Conventional DC Subsets Present in the Per-LNs in Steady State and during Inflammation

In steady state, per-LN migratory cDCs comprise epidermal LCs differentiated from monocytes and dermal DCs that have also been proposed to derive from monocytes. Both migratory cDC subsets constitutively migrate to the per-LNs and acquire a mature phenotype. Resident cDCs include $CD8^+$ and $CD8^-$ DCs that remain in an immature state in the absence of activating stimuli and differentiate from a common nonmonocytic precursor. During inflammatory reactions caused by infection, pre-existing LCs and dermal DCs rapidly migrate to the per-LNs and are replaced by newly formed LCs and dermal mo-DCs derived from inflammatory Ly-$6C^+$ monocytes recruited to the dermis. Dermal mo-DCs are phenotypically similar to dermal DCs and also migrate to the per-LNs as mature DCs. Whether newly formed LCs migrate to the per-LNs during infection remains to be demonstrated. Inflammatory monocytes are also recruited to the per-LNs and differentiate into mo-DCs. Finally, resident inflammatory $CD8^+$ and $CD8^-$ DCs are also formed during infection, but their precursors have yet to be defined.

after infection (Leon and Ardavin, 2008a; Martin et al., 2002; Serbina et al., 2003; Yoneyama et al., 2001), and the rapid recruitment of monocytes to inflammed foci (Henderson et al., 2003; Le Borgne et al., 2006; Palframan et al., 2001; Randolph et al., 1999; Serbina and Pamer, 2006), we can conclude that, during infection, migratory and resident cDCs existing before the onset of an infectious process are rapidly replaced by newly formed cDCs. These will include de novo-formed inflammatory mo-DCs, as well as $CD8^+$ and $CD8^-$ DCs. In this regard, studies addressing the role of defined cDC subsets during infection have generally been interpreted on the basis of previous information on migratory and resident cDCs preexisting in steady state. This could have led to misinterpretations due to possible functional differences between cDC subsets in steady state versus inflammatory conditions, such as differences in their migration behavior, antigen-handling capacity, and immunostimulatory potential. Consequently, a number of conflicting points have arisen in regards to de novo DC formation during infection. Are steady-state dermal DCs and inflammatory dermal mo-DCs equivalent? Are $CD8^+$ and $CD8^-$ DCs formed during inflammation functionally equivalent to their counterparts in steady state? Additionally, de novo formation of mo-DCs from monocytes recruited to the per-LNs or spleen has not generally been taken into consideration when the role of dermal DCs or resident $CD8^-$ DCs during infection has been assessed. Thus, in certain experimental infection models, a specific function that could be fulfilled by dermal mo-DCs or LN mo-DCs could have been ascribed erroneously to dermal DCs. Similarly, some functions that could be carried out by dermal, LN, or spleen mo-DCs could have been attributed incorrectly to $CD8^-$ DCs because in most reports exploring the function of $CD8^-$ DCs during infectious processes, $CD8^-$ DCs were characterized as $CD8^-$ $CD11b^+$, a definition that can also be applied to mo-DCs. These considerations should be taken into account in future studies aiming to ascertain the specific functions of defined DC subsets during infectious processes.

Functional Relevance of $CD8^+$ and $CD8^-$ DCs in T Cell Immunity

Reports published over the last decade point to a functional dichotomy in the role played by $CD8^+$ versus the role played by $CD8^-$ DCs in the induction of immune responses against pathogens. On the basis of these data, $CD8^+$ DCs were claimed to be specialized in the induction of anti-viral $CD8^+$ T cell responses, in part because of their crosspriming potential, whereas $CD8^-$ DCs were proposed to be mainly involved in $CD4^+$ T cell immunity,

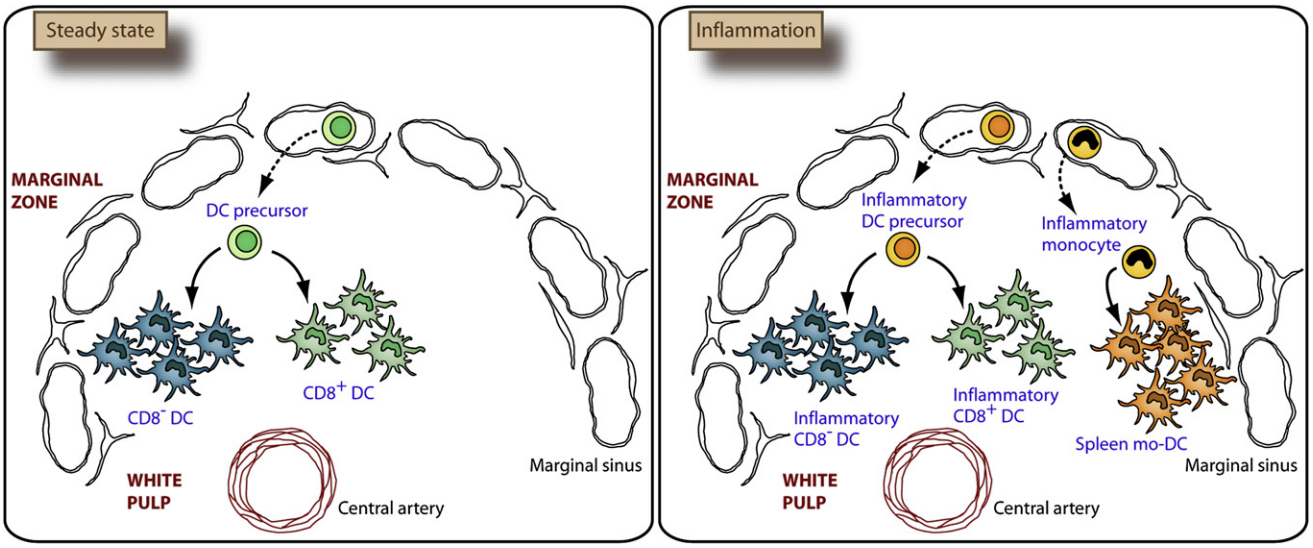

Figure 2. Conventional DC Subsets Present in the Spleen in Steady State and during Inflammation

In steady state, spleen resident cDCs comprise $CD8^+$ and $CD8^-$ DCs that, as described for their pre-LN counterparts, differentiate from a common nonmonocytic precursor and remain in an immature state in the absence of activating stimuli. During infection, inflammatory monocytes recruited to the splenic marginal zone differentiate locally into spleen mo-DCs, and pre-existing resident cDCs are replaced by de novo-formed inflammatory $CD8^+$ and $CD8^-$ DCs, derived from an undefined precursor.

particularly during bacterial infections (reviewed by Villadangos and Schnorrer, 2007). This concept has been further supported by a recent report proposing that the preferential ability of $CD8^+$ and $CD8^-$ DCs to prime $CD8^+$ and $CD4^+$ T cells, respectively, correlates with the differential capacity of these DC subsets to process and present antigens in association with MHC class I and MHC class II molecules, respectively (Dudziak et al., 2007). In addition, primarily on the basis of in vitro or combined in vivo-in vitro assays, $CD8^+$ and $CD8^-$ DCs were thought to induce T helper (Th) 1- and Th2-polarized responses, respectively (Maldonado-Lopez et al., 2001; Pulendran et al., 1999).

Table 1. Phenotype of Nonmucosal Conventional DCs

	$CD8^+$ DCs[a]	$CD8^-$ DCs[a]	LCs[a]	Dermal DCs[a]	Dermal mo-DCs[b]	LN mo-DCs[b]
CD11c	+++	+++	++	++	++	++
CD11b	+	+++	+++	++	++	+++
Ly-6C	–	–	ND	++	++	+++
CD8α	+++	–	++	++	++	–
SIRPα	+	+++	++	++	ND	ND
MHC II	++	++	+++	+++	+++	++
DEC-205	+++	–	+++	++	++	–
DCIR2	–	+++	–	–	ND	ND
Langerin	++	–	+++	–/+++[c]	ND	ND
CD103	++	–	–	++	ND	ND

"–," "+," "++," and "+++" correspond to null, low, intermediate, and high relative expression of the indicated marker, respectively, for the different DC subsets. ND, not determined.

[a] Phenotype in steady state for spleen and per-LN $CD8^+$ and $CD8^-$ DCs and for LCs and dermal DCs after migration into the per-LNs.

[b] Phenotype of mo-DCs generated in the dermis and per-LNs.

[c] Dermal DCs can be subdivided into $Langerin^+$ and $Langerin^-$ subsets.

However, recent studies involving the analysis of in vivo immune responses during infection have challenged these concepts by demonstrating that $CD8^+$ DCs are also involved in the induction $CD4^+$ and $CD8^+$ T cell responses, not only during viral infections, but also during bacterial and parasitic infections (Belz et al., 2005; Maroof and Kaye, 2008; Yarovinsky et al., 2006). In addition, under certain experimental conditions, $CD8^-$ DCs can produce interleukin-12 (IL-12) (Edwards et al., 2003; Fallarino et al., 2002; Fujii et al., 2003; Maldonado-Lopez et al., 2001) and could therefore be involved in the induction of Th1 cell responses against pathogens.

Although specific functions related to defense mechanisms against viruses, bacteria, and parasites have been attributed to different subsets of cDCs on the basis of recent advances in DC-mediated immunity against infection, there still are controversial points to be clarified. It is important to bear in mind that current available information on the functional specialization of DC subsets relies on studies that explored DC-mediated immune responses against a limited number of pathogens and that were performed with diverse experimental strategies. In addition, even though some precise functions have been assigned to individual DC subsets on the basis of the expression of C-type lectin antigen receptors (Carter et al., 2006; Dudziak et al., 2007; Sancho et al., 2008), TLR activation receptors (Yarovinsky et al., 2005), and chemokine receptors (Aliberti et al., 2000), a complete definition of specific expression profiles for these receptors and antigen-processing machinery should be performed to accurately delineate the role of each DC subset in defense against infection.

Roles of $CD8^+$ DCs and Dermal DCs in Immune Responses against Pathogens

In vivo studies that used mouse experimental models of infection by cytolytic and noncytolytic viruses (Allan et al., 2003; Allan

et al., 2006; Belz et al., 2004a; Belz et al., 2005; Dalod et al., 2002; Mount et al., 2008; Smith et al., 2003; Wilson et al., 2006), bacteria (Belz et al., 2005; Yrlid and Wick, 2002), and parasites (Sponaas et al., 2006; Yarovinsky et al., 2006) strongly support the concept that under physiological conditions, $CD8^+$ DCs fulfill a crucial role in the induction of protective Th1 cell responses against intracellular pathogens. Consistent with this concept, $CD8^+$ DCs have been shown to be extremely efficient in producing IL-12 in vivo after microbial stimulation (Maldonado-Lopez et al., 2001; Pulendran et al., 1999) and during in vivo infection (Dalod et al., 2002; Maroof and Kaye, 2008; Reis e Sousa et al., 1997; Yrlid and Wick, 2002). Interestingly, $CD8^+$ DCs have also been recently demonstrated to induce Th1 cell responses in vivo by an IL-12-independent, CD70-dependent mechanism (Soares et al., 2007).

$CD8^+$ DCs can activate $CD4^+$ and $CD8^+$ T cell responses after internalization of pathogens or pathogen-derived antigens in per-LNs or the spleen, either by direct presentation to $CD4^+$ or $CD8^+$ T cells or by crosspresentation to $CD8^+$ T cells (Allan et al., 2003; Allan et al., 2006; Belz et al., 2004a; Belz et al., 2005; Dalod et al., 2002; Mount et al., 2008; Smith et al., 2003; Sponaas et al., 2006; Wilson et al., 2006: Yarovinsky et al., 2006; Yrlid and Wick, 2002). Interestingly, recent data have led to the concept that during cutaneous infections, skin migrating dermal DCs and per-LN resident $CD8^+$ DCs cooperate for the induction of immune responses (Allan et al., 2003; Allan et al., 2006; Belz et al., 2004b). During viral infections, per-LN $CD8^+$ DCs have been demonstrated to activate $CD8^+$ T cells by cross-presentation of viral antigens transferred to $CD8^+$ DCs by dermal DCs that have uptaken those pathogens in the dermis and migrated to the per-LNs; these dermal DCs were responsible for the induction, within the per-LNs, of virus-specific $CD4^+$ T cells responses (Allan et al., 2006; Mount et al., 2008). In this regard, several reports support the idea that dermal DCs play an essential role in the induction of $CD4^+$ T cell responses against pathogens (Leon et al., 2007; Mount et al., 2008; Shklovskaya et al., 2008). Correspondingly, dermal DCs express different C-type lectin receptors, such as DEC-205, Langerin, and dectin-1, allowing them to internalize exogenous antigens efficiently (Carter et al., 2006; Ginhoux et al., 2007; Shklovskaya et al., 2008). A role for dermal DCs in the crosspriming of $CD8^+$ T cells during infection cannot be excluded, and such a role is supported by dermal DCs' expression of the C-type lectin DEC-205 (Shklovskaya et al., 2008) and data on the crosspriming capacity of dermal mo-DCs (Le Borgne et al., 2006).

$CD8^+$ DCs are highly efficient in processing and presenting antigens through the MHC class I pathway, either by direct presentation or crosspresentation (den Haan et al., 2000; Dudziak et al., 2007; Schnorrer et al., 2006), and $CD8^+$ DCs represent the main DC subset responsible for the induction of $CD8^+$ T cell responses against pathogens in vivo, particularly during viral infections (Allan et al., 2003; Allan et al., 2006; Belz et al., 2004a; Belz et al., 2005; Mount et al., 2008; Smith et al., 2003; Wilson et al., 2006). The high crosspriming efficiency of $CD8^+$ DCs has been correlated with their ability to internalize dying cell-associated antigens (Iyoda et al., 2002; Schulz and Reis e Sousa, 2002) and with the expression of C-type lectin antigen receptors, such as DEC-205 (Dudziak et al., 2007), CD36 (Tagliani et al., 2008), and the recently described DC, NK lectin group receptor-1 (DNGR-1; Sancho et al., 2008), that target antigens to the crosspriming processing pathway. Consistent with this notion, analysis of gene-expression profiles suggest that $CD8^+$ DCs, in contrast to $CD8^+$ DCs, are endowed with the antigen-processing machinery required for crosspresentation (Dudziak et al., 2007).

A crucial role for $CD8^+$ DCs in the induction of Th1 cell responses has also been demonstrated after infection by the enteroinvasive parasite *Toxoplama gondii* (Reis e Sousa et al., 1997). Interestingly, IL-12 production by $CD8^+$ DCs was triggered after recognition of *Toxoplasma*-derived profilin by TLR11, expressed by $CD8^+$ DCs (Yarovinsky et al., 2005). Splenic $CD8^+$ DCs efficiently uptake *Listeria monocytogenes* and appear to be essential for bacterial entry into the spleen and subsequent initiation of defense mechanisms against this microorganism (Neuenhahn et al., 2006). Finally, an immunoregulatory function leading to the suppression of T cell responses and the control of excessive inflammatory reactions has been proposed for $CD8^+$ DCs; this function depends on a mechanism mediated by the tryptophan metabolism pathway and initiated by the induction of the enzyme indoleamine 2,3-dioxygenase (IDO) (Fallarino et al., 2004). Interestingly, recent data support a role for regulatory T cells in the induction of IDO in $CD8^+$ DCs by a reverse signaling mechanism that triggers the noncanonical activation of the transcription factor NF-κB in $CD8^+$ DCs (reviewed by Puccetti and Grohmann, 2007). However, a direct implication of IDO-mediated negative regulation by $CD8^+$ DCs during in vivo immune responses against pathogens remains to be demonstrated.

Role of $CD8^-$ DCs in Immune Responses against Pathogens

In contrast to $CD8^+$ DCs, $CD8^-$ DCs are very effective in capturing pathogens and pathogen-derived antigens by phagocytosis and endocytosis and presenting them to $CD4^+$ T cells through the MHC class II pathway (reviewed by Villadangos and Schnorrer, 2007). In this regard, $CD8^-$ DCs efficiently induce $CD4^+$ T cell responses after antigen targeting via C-type lectin receptors, such as dectin-1 and DCIR-2 (Carter et al., 2006; Dudziak et al., 2007). However, $CD8^-$ DCs have neither the capacity to internalize apoptotic cells nor to crosspresent antigens to $CD8^+$ T cells (den Haan et al., 2000; Iyoda et al., 2002; Schulz and Reis e Sousa, 2002). $CD8^-$ DCs express the C-type lectin receptor DCIR2, recognized by the mAb 33D1, that has been proposed to target antigens to the endocytic route involved in antigen presentation to $CD4^+$ T cells in the context of MHC class II molecules. This pathway contrasts with the one involving DEC-205 that directs antigens to the processing pathway leading to antigen crosspresentation in the context of MHC class I molecules (Dudziak et al., 2007). Thus, it appears that genes related to the MHC class I processing and crosspresentation machinery were preferentially activated in $CD8^+$ DCs, whereas those related to the MHC class II processing machinery were preferentially activated in $CD8^-$ DCs. The inefficiency of $CD8^-$ DCs at crosspresenting antigens would therefore be due to the lack of the crosspresentation machinery and the lack of expression of C-type lectin receptors, such as DEC-205 or DNGR-1, that target antigens to the crosspresentation pathway.

As mentioned above, data on the induction of polarized Th1 and Th2 cell responses by $CD8^+$ and $CD8^-$ DCs against soluble

antigens originally led to the notion that $CD8^-$ DCs were poor IL-12 producers and were mainly involved in the induction of Th2 cell responses (Maldonado-Lopez et al., 2001; Pulendran et al., 1999). In line with this proposal, in several experimental models of infection by viruses, bacteria, and parasites, $CD8^+$ DCs, but not $CD8^-$ DCs, were shown to be responsible for IL-12 production and induction of Th1 cell responses (Dalod et al., 2002; Maroof and Kaye, 2008; Reis e Sousa et al., 1997; Yrlid and Wick, 2002). In addition, $CD8^-$ DCs have been reported to activate Th2-polarized T cell responses in vivo during infection from *Plasmodium chabaudi* malaria parasites (Sponaas et al., 2006). However, under certain experimental conditions, $CD8^-$ DCs can produce IL-12. Thus, IL-12 production by $CD8^-$ DCs was demonstrated in response to TLR-7 ligands (Edwards et al., 2003), α-galactosylceramide (Fujii et al., 2003), and LPS in the presence of anti-IL-10 (Fallarino et al., 2002; Maldonado-Lopez et al., 2001). Interestingly, splenic $CD8^-$ DCs have been demonstrated to induce $CD4^+$ T cell responses during influenza virus infection (Mount et al., 2008) and to induce Th1 cell-biased OVA-specific T cell responses after LPS stimulation by a mechanism that relies on Delta-4 Notch-ligand signaling and is independent of IL-12 production (Skokos and Nussenzweig, 2007).

These data suggest that $CD8^-$ DCs could participate in either Th1 or Th2 cell responses depending on different factors, such as the activating stimulus and the internalization, processing, and presentation of pathogen-derived antigens. Unfortunately, only a limited number of reports analyzing the role of $CD8^-$ DCs in immunity against pathogens have been published, and consequently, additional experiments should be performed to improve our understanding of the role played by $CD8^-$ DCs in immunity.

$CD8^-$ DCs have also been proposed to have a microbicidal function through the production TNF-α and iNOS (Serbina et al., 2003) and to participate in the regulation of Ig production by B cells (Balazs et al., 2002). However, on the basis of recent information on inflammatory de novo-formed DCs, it appears that these functions could have been incorrectly attributed to $CD8^-$ DCs and could be fulfilled by mo-DCs, as discussed in the next section.

$CD8^-$ DCs express the inhibitory receptor SIRPα (Lahoud et al., 2006), which appears to negatively control LC migration (Fukunaga et al., 2004) and has been demonstrated to negatively control IL-12 production after interaction with T cells expressing SIPRα counter-receptor CD47 in human DCs (Latour et al., 2001). This receptor has a crucial role in $CD8^-$ DC development because this DC subset is severely reduced in CD47-deficient mice (Hagnerud et al., 2006).

Role of Inflammatory mo-DCs in Immune Responses against Pathogens

mo-DCs differentiated in the skin, per-LNs, and spleen during infectious processes can play an important role in the induction and regulation of immune responses against pathogens (reviewed in Leon and Ardavin, 2008b). Although only a few reports have specifically addressed the role of mo-DCs in innate and adaptive immunity, these studies have demonstrated that mo-DCs can be essential for the induction of effective defense mechanisms against infection. This process applies particularly to nonacute infectious and inflammatory processes that involve the replacement of pre-existing DC subsets by de novo-formed inflammatory DCs. It is especially important to take into consideration that, as discussed above, some relevant functions that have been assigned to dermal DCs could be actually fulfilled by dermal mo-DCs. These functions could include the capture and transport of antigens to per-LN $CD8^+$ DC for crosspresentation, the induction of $CD4^+$ T cell responses by direct priming, and, conceivably, $CD8^+$ T cell crosspriming. This concept is supported first by the fact that within the per-LNs, both DC subsets have a similar, if not identical, phenotype, and second by the kinetics of dermal DC migration and de novo dermal mo-DC differentiation, which suggests that dermal DCs are rapidly replaced by dermal mo-DCs after the onset of an infectious process.

In line with these considerations, during *Leishmania major* infection, monocytes were recruited to the dermis, draining per-LNs, and spleen and differentiated into dermal mo-DCs, LN mo-DCs, and spleen mo-DCs, respectively (Leon et al., 2007). Dermal mo-DCs acquired a mature phenotype after migration to the per-LNs and were demonstrated to be responsible for the induction of protective $CD4^+$ Th1 cell responses against the parasite. Consistent with this finding, during subcutaneous *Salmonella* infection, $CD4^+$ T cells were activated by DCs derived from monocytes recruited to the dermis by a CCR6-dependent mechanism (Ravindran et al., 2007). In contrast, LN mo-DCs differentiated from monocytes recruited to the per-LNs during *Leishmania* infection (Leon et al., 2007) exhibited an immature phenotype and did not appear to contribute substantially to T cell immunity against *Leishmania*. The function of LN mo-DCs and spleen mo-DCs during *Leishmania* infection remains to be defined, although it can be speculated that they could participate in innate-immunity defense mechanisms, on the basis of the role of spleen mo-DCs during *Listeria* infection (Serbina et al., 2003). Splenic DCs, formed de novo during infection by *Listeria monocytogenes* from monocytes recruited to the spleen by a CCR2-dependent mechanism, displayed a highly effective microbicidal potential on the basis of their capacity to produce TNF-α and nitric oxide; these mo-DCs have been named Tip DCs (TNF-α- and iNOS-producing DCs). Tip DCs, which displayed a similar phenotype to LN and spleen mo-DCs formed in *Leishmania*-infected mice (Leon et al., 2007), were essential for defense against *Listeria* infection, as demonstrated in CCR2-deficient mice that could not control bacterial replication (Serbina et al., 2003). Interestingly, DCs newly formed in the marginal zone of the spleen during infection by *Streptococcus pneumoniae* were also claimed to correspond phenotypically to Tip DCs (Balazs et al., 2002). These DCs were involved in the induction of T cell-independent B cell responses and in the differentiation of IgM-producing plasma cells by a mechanism relying on the production, by DCs, of the recently described TNF superfamily molecules B lymphocyte stimulator (BlyS) and a proliferation-inducing ligand (APRIL).

These data suggest that whereas mature dermal mo-DCs display an essential role in the induction of pathogen-specific T cell responses, immature LN and spleen mo-DCs would be involved in pathogen killing and participate in the regulation of Ig production by B cells.

Conclusions and Future Research Directions

Recent research on the specific functions fulfilled by different DC subsets during in vivo immune responses have challenged

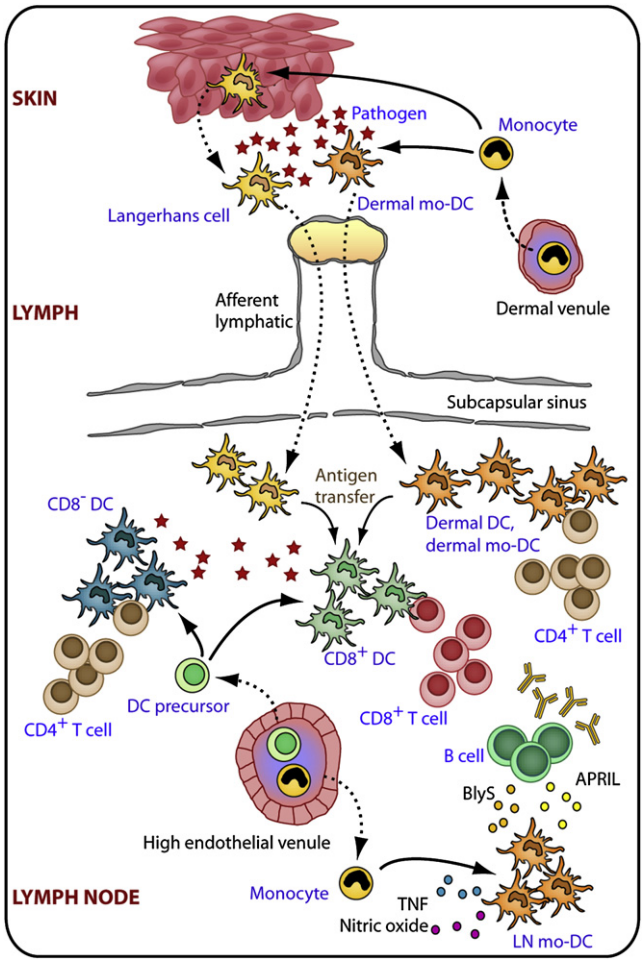

Figure 3. Integrated Model of the Specific Functions Ascribed to the Different Per-LN cDC Subsets during Infectious Processes

Migratory monocyte-derived LCs, dermal DCs, and dermal mo-DCs capture pathogens or pathogen-derived antigens in the skin, transport them to the pre-LNs, and can transfer them to resident $CD8^+$ DCs. Both dermal DCs and dermal mo-DCs can also efficiently activate $CD4^+$ T cells within the per-LNs. $CD8^+$ DCs are highly efficient in activating $CD8^+$ T cells by direct presentation or crosspresentation, whereas resident $CD8^-$ DCs would preferentially activate CD4+ T cells. Finally, LN mo-DCs that display an immature phenotype can have a microbicidal function by producing TNFα and nitric oxide, and they participate in Ig production by B cells through a mechanism dependent on the production of APRIL and BlyS by mo-DCs.

certain paradigms of DC immunobiology, revealed unexpected functions for some DC subsets, and contributed to the description of new DC subsets. Whereas the participation of LCs in immunity against pathogens under physiological conditions represents a yet-unsolved controversy, dermal DCs have recently arisen as critical effectors of immune responses during cutaneous infections by virtue of their capacity to efficiently capture, transport, and present pathogen-derived antigens to per-LN $CD4^+$ T cells. Moreover, they enable the crosspriming of $CD8^+$ T cells by resident $CD8^+$ DCs by providing the latter with pathogen-derived antigens. Importantly, the finding that mo-DCs formed de novo during infection in antigen-capture areas and lymphoid organs can be responsible for the induction of protective immune responses against pathogens has led to the development of a new active area of research in DC immunobiology, one that will provide important insights regarding the induction and control of immune responses during inflammation and infection (see Belkaid and Oldenhove, 2008 [in this issue]). An integrated view of the specific functions ascribed to migratory and resident cDCs during infection is shown in Figure 3.

There are numerous controversial and unresolved issues surrounding DC-mediated immune responses against pathogens. Therefore, definitive conclusions cannot yet be drawn on the complete set of functions fulfilled by each cDC subset, functions that appear to be defined by multiple factors, including the control of their differentiation under steady state and inflammation, their location within lymphoid organs, and the expression of specific antigen-capture receptors, TLR and other activating receptors, cytokine and chemokine receptors, and components of the antigen-processing and presentation machinery. Therefore, an in-depth analysis of all these aspects of the immunobiology of the different DC subsets would contribute substantially to the defining of the functional specialization of the different cDC subsets during immune responses against pathogens.

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REFERENCES

Aliberti, J., Reis e Sousa, C., Schito, M., Hieny, S., Wells, T., Huffnagle, G.B., and Sher, A. (2000). CCR5 provides a signal for microbial induced production of IL-12 by CD8 alpha+ dendritic cells. Nat. Immunol. *1*, 83–87.

Allan, R.S., Smith, C.M., Belz, G.T., van Lint, A.L., Wakim, L.M., Heath, W.R., and Carbone, F.R. (2003). Epidermal viral immunity induced by CD8alpha+ dendritic cells but not by Langerhans cells. Science *301*, 1925–1928.

Allan, R.S., Waithman, J., Bedoui, S., Jones, C.M., Villadangos, J.A., Zhan, Y., Lew, A.M., Shortman, K., Heath, W.R., and Carbone, F.R. (2006). Migratory dendritic cells transfer antigen to a lymph node-resident dendritic cell population for efficient CTL priming. Immunity *25*, 153–162.

Aoshi, T., Zinselmeyer, B.H., Konjufca, V., Lynch, J.N., Zhang, X., Koide, Y., and Miller, M.J. (2008). Bacterial entry to the splenic white pulp initiates antigen presentation to $CD8^+$ T cells. Immunity *29*, this issue, 476–486.

Balazs, M., Martin, F., Zhou, T., and Kearney, J. (2002). Blood dendritic cells interact with splenic marginal zone B cells to initiate T-independent immune responses. Immunity *17*, 341–352.

Belkaid, Y., and Oldenhove, G. (2008). Tuning microenvironments: Induction of regulatory T cells by dendritic cells. Immunity *29*, this issue, 362–371.

Belz, G.T., Shortman, K., Bevan, M.J., and Heath, W.R. (2005). CD8alpha+ dendritic cells selectively present MHC class I-restricted noncytolytic viral and intracellular bacterial antigens in vivo. J. Immunol. *175*, 196–200.

Belz, G.T., Smith, C.M., Eichner, D., Shortman, K., Karupiah, G., Carbone, F.R., and Heath, W.R. (2004a). Cutting edge: Conventional CD8 alpha+ dendritic cells are generally involved in priming CTL immunity to viruses. J. Immunol. *172*, 1996–2000.

Belz, G.T., Smith, C.M., Kleinert, L., Reading, P., Brooks, A., Shortman, K., Carbone, F.R., and Heath, W.R. (2004b). Distinct migrating and nonmigrating dendritic cell populations are involved in MHC class I-restricted antigen presentation after lung infection with virus. Proc. Natl. Acad. Sci. USA *101*, 8670–8675.

Carter, R.W., Thompson, C., Reid, D.M., Wong, S.Y., and Tough, D.F. (2006). Preferential induction of CD4+ T cell responses through in vivo targeting of antigen to dendritic cell-associated C-type lectin-1. J. Immunol. *177*, 2276–2284.

Dalod, M., Salazar-Mather, T.P., Malmgaard, L., Lewis, C., Asselin-Paturel, C., Brière, F., Trinchieri, G., and Biron, C.A. (2002). Interferon alpha/beta and interleukin 12 responses to viral infections: Pathways regulating dendritic cell cytokine expression in vivo. J. Exp. Med. *195*, 517–528.

de Heer, H.J., Hammad, H., Kool, M., and Lambrecht, B.N. (2005). Dendritic cell subsets and immune regulation in the lung. Semin. Immunol. *17*, 295–303.

De Smedt, T., Pajak, B., Muraille, E., Lespagnard, L., Heinen, E., De Baetselier, P., Urbain, J., Leo, O., and Moser, M. (1996). Regulation of dendritic cell numbers and maturation by lipopolysaccharide in vivo. J. Exp. Med. *184*, 1413–1424.

den Haan, J.M., Lehar, S.M., and Bevan, M.J. (2000). CD8(+) but not CD8(-) dendritic cells cross-prime cytotoxic T cells in vivo. J. Exp. Med. *192*, 1685–1696.

Dudziak, D., Kamphorst, A.O., Heidkamp, G.F., Buchholz, V.R., Trumpfheller, C., Yamazaki, S., Cheong, C., Liu, K., Lee, H.W., Park, C.G., et al. (2007). Differential antigen processing by dendritic cell subsets in vivo. Science *315*, 107–111.

Edwards, A.D., Diebold, S.S., Slack, E.M., Tomizawa, H., Hemmi, H., Kaisho, T., Akira, S., and Reis e Sousa, C. (2003). Toll-like receptor expression in murine DC subsets: Lack of TLR7 expression by CD8 alpha+ DC correlates with unresponsiveness to imidazoquinolines. Eur. J. Immunol. *33*, 827–833.

Fallarino, F., Asselin-Paturel, C., Vacca, C., Bianchi, R., Gizzi, S., Fioretti, M.C., Trinchieri, G., Grohmann, U., and Puccetti, P. (2004). Murine plasmacytoid dendritic cells initiate the immunosuppressive pathway of tryptophan catabolism in response to CD200 receptor engagement. J. Immunol. *173*, 3748–3754.

Fallarino, F., Grohmann, U., Vacca, C., Bianchi, R., Fioretti, M.C., and Puccetti, P. (2002). CD40 ligand and CTLA-4 are reciprocally regulated in the Th1 cell proliferative response sustained by CD8(+) dendritic cells. J. Immunol. *169*, 1182–1188.

Filippi, C., Hugues, S., Cazareth, J., Julia, V., Glaichenhaus, N., and Ugolini, S. (2003). CD4+ T cell polarization in mice is modulated by strain-specific major histocompatibility complex-independent differences within dendritic cells. J. Exp. Med. *198*, 201–209.

Fujii, S., Shimizu, K., Smith, C., Bonifaz, L., and Steinman, R.M. (2003). Activation of natural killer T cells by alpha-galactosylceramide rapidly induces the full maturation of dendritic cells in vivo and thereby acts as an adjuvant for combined CD4 and CD8 T cell immunity to a coadministered protein. J. Exp. Med. *198*, 267–279.

Fukunaga, A., Nagai, H., Noguchi, T., Okazawa, H., Matozaki, T., Yu, X., Lagenaur, C.F., Honma, N., Ichihashi, M., Kasuga, M., et al. (2004). Src homology 2 domain-containing protein tyrosine phosphatase substrate 1 regulates the migration of Langerhans cells from the epidermis to draining lymph nodes. J. Immunol. *172*, 4091–4099.

Ginhoux, F., Tacke, F., Angeli, V., Bogunovic, M., Loubeau, M., Dai, X.M., Stanley, E.R., Randolph, G.J., and Merad, M. (2006). Langerhans cells arise from monocytes in vivo. Nat. Immunol. *7*, 265–273.

Ginhoux, F., Collin, M.P., Bogunovic, M., Abel, M., Leboeuf, M., Helft, J., Ochando, J., Kissenpfennig, A., Malissen, B., Grisotto, M., et al. (2007). Blood-derived dermal langerin+ dendritic cells survey the skin in the steady state. J. Exp. Med. *204*, 3133–3146.

Hagnerud, S., Manna, P.P., Cella, M., Stenberg, A., Frazier, W.A., Colonna, M., and Oldenborg, P.A. (2006). Deficit of CD47 results in a defect of marginal zone dendritic cells, blunted immune response to particulate antigen and impairment of skin dendritic cell migration. J. Immunol. *176*, 5772–5778.

Henderson, R.B., Hobbs, J.A., Mathies, M., and Hogg, N. (2003). Rapid recruitment of inflammatory monocytes is independent of neutrophil migration. Blood *102*, 328–335.

Iwasaki, A. (2007). Mucosal dendritic cells. Annu. Rev. Immunol. *25*, 381–418.

Iyoda, T., Shimoyama, S., Liu, K., Omatsu, Y., Akiyama, Y., Maeda, Y., Takahara, K., Steinman, R.M., and Inaba, K. (2002). The CD8+ dendritic cell subset selectively endocytoses dying cells in culture and in vivo. J. Exp. Med. *195*, 1289–1302.

Johansson, C., and Kelsall, B.L. (2005). Phenotype and function of intestinal dendritic cells. Semin. Immunol. *17*, 284–294.

Kamath, A.T., Henri, S., Battye, F., Tough, D.F., and Shortman, K. (2002). Developmental kinetics and lifespan of dendritic cells in mouse lymphoid organs. Blood *100*, 1734–1741.

Kissenpfennig, A., and Malissen, B. (2006). Langerhans cells–revisiting the paradigm using genetically engineered mice. Trends Immunol. *27*, 132–139.

Lahoud, M.H., Proietto, A.I., Gartlan, K.H., Kitsoulis, S., Curtis, J., Wettenhall, J., Sofi, M., Daunt, C., O'Keeffe, M., Caminschi, I., et al. (2006). Signal regulatory protein molecules are differentially expressed by CD8- dendritic cells. J. Immunol. *177*, 372–382.

Latour, S., Tanaka, H., Demeure, C., Mateo, V., Rubio, M., Brown, E.J., Maliszewski, C., Lindberg, F.P., Oldenborg, A., Ullrich, A., et al. (2001). Bidirectional negative regulation of human T and dendritic cells by CD47 and its cognate receptor signal-regulator protein-alpha: Down-regulation of IL-12 responsiveness and inhibition of dendritic cell activation. J. Immunol. *167*, 2547–2554.

Le Borgne, M., Etchart, N., Goubier, A., Lira, S.A., Sirard, J.C., van Rooijen, N., Caux, C., Ait-Yahia, S., Vicari, A., Kaiserlian, D., and Dubois, B. (2006). Dendritic cells rapidly recruited into epithelial tissues via CCR6/CCL20 are responsible for CD8+ T cell crosspriming in vivo. Immunity *24*, 191–201.

Lemos, M.P., Esquivel, F., Scott, P., and Laufer, T.M. (2004). MHC class II expression restricted to CD8alpha+ and CD11b+ dendritic cells is sufficient for control of Leishmania major. J. Exp. Med. *199*, 725–730.

Leon, B., and Ardavin, C. (2008a). Monocyte migration to inflamed skin and lymph nodes is differentially controlled by L-selectin and PSGL-1. Blood *111*, 3126–3130.

Leon, B., and Ardavin, C. (2008b). Monocyte-derived dendritic cells in innate and adaptive immunity. Immunol. Cell Biol. *86*, 320–324.

Leon, B., Lopez-Bravo, M., and Ardavin, C. (2007). Monocyte-derived dendritic cells formed at the infection site control the induction of protective T helper 1 responses against Leishmania. Immunity *26*, 519–531.

Leon, B., Martinez del Hoyo, G., Parrillas, V., Vargas, H.H., Sanchez-Mateos, P., Longo, N., Lopez-Bravo, M., and Ardavin, C. (2004). Dendritic cell differentiation potential of mouse monocytes: Monocytes represent immediate precursors of CD8- and CD8+ splenic dendritic cells. Blood *103*, 2668–2676.

Maldonado-Lopez, R., Maliszewski, C., Urbain, J., and Moser, M. (2001). Cytokines regulate the capacity of CD8alpha(+) and CD8alpha(-) dendritic cells to prime Th1/Th2 cells in vivo. J. Immunol. *167*, 4345–4350.

Maroof, A., and Kaye, P.M. (2008). Temporal regulation of interleukin-12p70 (IL-12p70) and IL-12-related cytokines in splenic dendritic cell subsets during Leishmania donovani infection. Infect. Immun. *76*, 239–249.

Martin, P., Ruiz, S.R., del Hoyo, G.M., Anjuere, F., Vargas, H.H., Lopez-Bravo, M., and Ardavin, C. (2002). Dramatic increase in lymph node dendritic cell number during infection by the mouse mammary tumor virus occurs by a CD62L-dependent blood-borne DC recruitment. Blood *99*, 1282–1288.

Mount, A.M., Smith, C.M., Kupresanin, F., Stoermer, K., Heath, W.R., and Belz, G.T. (2008). Multiple dendritic cell populations activate CD4+ T cells after viral stimulation. PLoS ONE *3*, e1691.

Naik, S.H., Metcalf, D., van Nieuwenhuijze, A., Wicks, I., Wu, L., O'Keeffe, M., and Shortman, K. (2006). Intrasplenic steady-state dendritic cell precursors that are distinct from monocytes. Nat. Immunol. *7*, 663–671.

Neuenhahn, M., Kerksiek, K.M., Nauerth, M., Suhre, M.H., Schiemann, M., Gebhardt, F.E., Stemberger, C., Panthel, K., Schroder, S., Chakraborty, T., et al. (2006). CD8alpha+ dendritic cells are required for efficient entry of Listeria monocytogenes into the spleen. Immunity *25*, 619–630.

Palframan, R.T., Jung, S., Cheng, G., Weninger, W., Luo, Y., Dorf, M., Littman, D.R., Rollins, B.J., Zweerink, H., Rot, A., and von Andrian, U.H. (2001). Inflammatory chemokine transport and presentation in HEV: A remote control mechanism for monocyte recruitment to lymph nodes in inflamed tissues. J. Exp. Med. *194*, 1361–1373.

Puccetti, P., and Grohmann, U. (2007). IDO and regulatory T cells: A role for reverse signalling and non-canonical NF-kappaB activation. Nat. Rev. Immunol. *7*, 817–823.

Pulendran, B., Smith, J.L., Caspary, G., Brasel, K., Pettit, D., Maraskovsky, E., and Maliszewski, C.R. (1999). Distinct dendritic cell subsets differentially regulate the class of immune response in vivo. Proc. Natl. Acad. Sci. USA *96*, 1036–1041.

Randolph, G.J., Inaba, K., Robbiani, D.F., Steinman, R.M., and Muller, W.A. (1999). Differentiation of phagocytic monocytes into lymph node dendritic cells in vivo. Immunity *11*, 753–761.

Ravindran, R., Rusch, L., Itano, A., Jenkins, M.K., and McSorley, S.J. (2007). CCR6-dependent recruitment of blood phagocytes is necessary for rapid CD4 T cell responses to local bacterial infection. Proc. Natl. Acad. Sci. USA *104*, 12075–12080.

Reis e Sousa, C., Hieny, S., Scharton-Kersten, T., Jankovic, D., Charest, H., Germain, R.N., and Sher, A. (1997). In vivo microbial stimulation induces rapid CD40 ligand-independent production of interleukin 12 by dendritic cells and their redistribution to T cell areas. J. Exp. Med. *186*, 1819–1829.

Sancho, D., Mourao-Sa, D., Joffre, O.P., Schulz, O., Rogers, N.C., Pennington, D.J., Carlyle, J.R., and Reis e Sousa, C. (2008). Tumor therapy in mice via antigen targeting to a novel, DC-restricted C-type lectin. J. Clin. Invest. *118*, 2098–2110.

Schnorrer, P., Behrens, G.M., Wilson, N.S., Pooley, J.L., Smith, C.M., El-Sukkari, D., Davey, G., Kupresanin, F., Li, M., Maraskovsky, E., et al. (2006). The dominant role of CD8+ dendritic cells in cross-presentation is not dictated by antigen capture. Proc. Natl. Acad. Sci. USA *103*, 10729–10734.

Schulz, O., and Reis e Sousa, C. (2002). Cross-presentation of cell-associated antigens by CD8alpha+ dendritic cells is attributable to their ability to internalize dead cells. Immunology *107*, 183–189.

Serbina, N.V., Salazar-Mather, T.P., Biron, C.A., Kuziel, W.A., and Pamer, E.G. (2003). TNF/iNOS-producing dendritic cells mediate innate immune defense against bacterial infection. Immunity *19*, 59–70.

Serbina, N.V., and Pamer, E.G. (2006). Monocyte emigration from bone marrow during bacterial infection requires signals mediated by chemokine receptor CCR2. Nat. Immunol. *7*, 311–317.

Shklovskaya, E., Roediger, B., and Fazekas de St Groth, B. (2008). Epidermal and dermal dendritic cells display differential activation and migratory behavior while sharing the ability to stimulate CD4+ T cell proliferation in vivo. J. Immunol. *181*, 418–430.

Shortman, K., and Naik, S.H. (2007). Steady-state and inflammatory dendritic-cell development. Nat. Rev. Immunol. *7*, 19–30.

Skokos, D., and Nussenzweig, M.C. (2007). CD8- DCs induce IL-12-independent Th1 differentiation through Delta 4 Notch-like ligand in response to bacterial LPS. J. Exp. Med. *204*, 1525–1531.

Smith, C.M., Belz, G.T., Wilson, N.S., Villadangos, J.A., Shortman, K., Carbone, F.R., and Heath, W.R. (2003). Conventional CD8 alpha+ dendritic cells are preferentially involved in CTL priming after footpad infection with herpes simplex virus-1. J. Immunol. *170*, 4437–4440.

Soares, H., Waechter, H., Glaichenhaus, N., Mougneau, E., Yagita, H., Mizenina, O., Dudziak, D., Nussenzweig, M.C., and Steinman, R.M. (2007). A subset of dendritic cells induces CD4+ T cells to produce IFN-gamma by an IL-12-independent but CD70-dependent mechanism in vivo. J. Exp. Med. *204*, 1095–1106.

Sponaas, A.M., Cadman, E.T., Voisine, C., Harrison, V., Boonstra, A., O'Garra, A., and Langhorne, J. (2006). Malaria infection changes the ability of splenic dendritic cell populations to stimulate antigen-specific T cells. J. Exp. Med. *203*, 1427–1433.

Steinman, R.M., Hawiger, D., and Nussenzweig, M.C. (2003). Tolerogenic dendritic cells. Annu. Rev. Immunol. *21*, 685–711.

Tagliani, E., Guermonprez, P., Sepulveda, J., Lopez-Bravo, M., Ardavin, C., Amigorena, S., Benvenuti, F., and Burrone, O.R. (2008). Selection of an antibody library identifies a pathway to induce immunity by targeting CD36 on steady-state CD8alpha+ dendritic cells. J. Immunol. *180*, 3201–3209.

Villadangos, J.A., and Heath, W.R. (2005). Life cycle, migration and antigen presenting functions of spleen and lymph node dendritic cells: Limitations of the Langerhans cells paradigm. Semin. Immunol. *17*, 262–272.

Villadangos, J.A., and Schnorrer, P. (2007). Intrinsic and cooperative antigen-presenting functions of dendritic-cell subsets in vivo. Nat. Rev. Immunol. *7*, 543–555.

Wilson, N.S., Behrens, G.M., Lundie, R.J., Smith, C.M., Waithman, J., Young, L., Forehan, S.P., Mount, A., Steptoe, R.J., Shortman, K.D., et al. (2006). Systemic activation of dendritic cells by Toll-like receptor ligands or malaria infection impairs cross-presentation and antiviral immunity. Nat. Immunol. *7*, 165–172.

Yarovinsky, F., Kanzler, H., Hieny, S., Coffman, R.L., and Sher, A. (2006). Toll-like receptor recognition regulates immunodominance in an antimicrobial CD4+ T cell response. Immunity *25*, 655–664.

Yarovinsky, F., Zhang, D., Andersen, J.F., Bannenberg, G.L., Serhan, C.N., Hayden, M.S., Hieny, S., Sutterwala, F.S., Flavell, R.A., Ghosh, S., and Sher, A. (2005). TLR11 activation of dendritic cells by a protozoan profilin-like protein. Science *308*, 1626–1629.

Yoneyama, H., Matsuno, K., Zhang, Y., Murai, M., Itakura, M., Ishikawa, S., Hasegawa, G., Naito, M., Asakura, H., and Matsushima, K. (2001). Regulation by chemokines of circulating dendritic cell precursors, and the formation of portal tract-associated lymphoid tissue, in a granulomatous liver disease. J. Exp. Med. *193*, 35–49.

Yrlid, U., and Wick, M.J. (2002). Antigen presentation capacity and cytokine production by murine splenic dendritic cell subsets upon Salmonella encounter. J. Immunol. *169*, 108–116.

Zhao, X., Deak, E., Soderberg, K., Linehan, M., Spezzano, D., Zhu, J., Knipe, D.M., and Iwasaki, A. (2003). Vaginal submucosal dendritic cells, but not Langerhans cells, induce protective Th1 responses to herpes simplex virus-2. J. Exp. Med. *197*, 153–162.