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Review

Dendritic cells and liver fibrosis ☆

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ABSTRACT

Dendritic cells are a relative rare population of specialized antigen presenting cells that are distributed through most lymphoid and non-lymphoid tissues and play a critical role in linking the innate and adaptive arms of the immune system. The liver contains a heterogeneous population of dendritic cells that may contribute to liver inflammation and fibrosis through a number of mechanisms. This review summarizes current knowledge on the development and characterization of liver dendritic cells and their potential impact on liver fibrosis. This article is part of a Special Issue entitled: Fibrosis: Translation of basic research to human disease.

1. Introduction

Dendritic cells (DCs) comprise a relative rare and heterogeneous population of specialized hematopoietic cells that play an important role in linking the innate and adaptive arms of the immune system. DCs were first identified as potent antigen presenting cells in mouse spleen and it is now established that they are distributed through most lymphoid and non-lymphoid tissues. Despite the fact that they typically represent only a small proportion within the leukocyte population, they are the primary professional antigen presenting cells and play an important role in monitoring the tissue microenvironment. After capturing antigens, tissue-resident DCs mature and migrate via the afferent lymphatics to the draining lymph nodes where they can present these antigens to T cells. The effectiveness of DCs at presenting antigens and priming T cell responses is one of their defining attributes. Their migratory capacity is also an important feature that distinguishes them from tissue-resident macrophages. DCs are also important producers of multiple cytokines through which they can influence a broad range of other cell types. DCs in the liver are uniquely positioned to monitor the portal circulation, and the ways in which they regulate responses to blood-borne pathogens, hepatic immune tolerance, liver homeostasis and fibrosis continue to be areas of active research. Given their scarcity, heterogeneity and the absence of clear defining surface markers, the investigation of hepatic DCs has thus far been challenging.

2. Subsets of liver dendritic cells

Tissue-resident DCs are present in most tissues; however, there is considerable functional and phenotypic heterogeneity amongst DC populations [1], which also varies based on tissue localization. The functional roles of DCs are often influenced by their specific tissue microenvironments, and certain tissues contain specialized DC populations, such as Langerhans cells in the skin. The DC populations in the liver express similar surface markers to those found in other tissues such as the lung, kidney and pancreas, and much of our understanding of liver DCs is based on studies of analogous DC populations that have been more extensively studied in other lymphoid and non-lymphoid tissues [2,3]. However, the liver seems to play a unique role in the DC traffic: at least in rat, DCs undergo a blood-lymph node translocation via the hepatic sinusoids, which may act as a biological concentrator of circulating DCs into the regional hepatic nodes [4].

DCs are sparsely distributed through the liver, and immunohistochemical studies of patient liver biopsies indicate that they are primarily found in the portal regions and occasionally in the parenchyma [5]. Unlike hematopoietic lineages such as B cells or T cells, a single cell surface marker that can be used to unequivocally identify DCs has yet to

Abbreviations: DC, dendritic cell; MHCII, major histocompatibility class II; IFN, interferon; CLP, common lymphoid progenitor; CMP, common myeloid progenitor; MDP, monocyte DC progenitor; CDP, common DC progenitor; Flt3, FMS-like tyrosine kinase 3; Flk2, fetal liver kinase-2; IRF8, interferon regulatory factor 8; zbtb4/zDC, zinc finger and BTB domain containing 4; ID-2, inhibitor of DNA binding 2; Batf3, basic leucine zipper transcriptional factor ATF-like 3; NK, natural killer; MMP, matrix metalloproteinase; BDL, bile duct ligation; MCP-1, monocyte chemotactic protein 1; TLR, toll-like receptor; DT, diphtheria toxin; TNFα, tumor necrosis factor alpha

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be determined, and distinguishing DCs from other cell types, such as monocytes and macrophages continues to be a challenge in some circumstances. The fact that some of the surface markers that are highly expressed by specific DC subsets in mice are poorly expressed on human DCs and vice versa presents an additional challenge. Nevertheless, considerable progress has been made in recent years in defining sets of cell surface markers that can be used to identify distinct DC subsets.

In multiparametric flow cytometric analyses, DCs in both mice and humans can be identified as $CD45^+$ cells that constitutively express high levels of major histocompatibility complex II (MHCII) while lacking markers for other hematopoietic lineages. This is admittedly a broad definition, and the use of additional markers, such as the high expression of CD11c, can be also useful in identifying DCs. However, effectively excluding other hematopoietic cell types when conducting DC analyses is very important given that many of the surface markers that frequently are used to define DC subsets can also be expressed by other cell types, such as B cells and macrophages. This is particularly true in the setting of inflammation.

Under steady state conditions, liver DCs in both mice and humans can be divided into two major functional classes: classical DCs (cDCs) and plasmacytoid DCs (pDCs). cDCs express high levels of MHCII and function as highly-efficient professional antigen presenting cells. In contrast, pDCs express relatively lower levels of MHCII and have a relatively limited capacity to capture and present tissue antigens and instead function as major producers of type I interferons (IFNs) in response to nucleic acids in the setting of viral infection [6] (Table 1). Reinforcing these functional differences, pDCs share certain molecular and morphological features with B lymphocytes, including typical secretory lymphocyte morphology rather than the eponymous dendritic morphology of cDCs. In naïve C57BL/B6 mice, pDCs comprise the majority of liver DCs and can be identified as a $CD11c^+$ population that expresses CD317 (PDCA1) and Siglec H [7] (Fig. 1A). Human pDCs do not express CD11c but can be identified as HLA-DR^{hi} cells that also express high levels of the type II C-type lectin CD303 (BDCA2) and the IL-3 receptor CD123 [8]. The frequency of these cells in human liver explants and resections is typically much lower than the frequency of pDCs found in the livers of naïve mice (Fig. 1B), and is similar to the frequency observed amongst circulating PBMCs.

The cDC population in the liver can be further divided into two major functionally and phenotypically distinct subsets. In mice, the hepatic $CD11c^{hi}MHCII^{hi}$ cDC population contains a more prevalent $CD103^-CD11b^{hi}$ population and a rarer $CD103^+CD11b^{low}$ population (Fig. 1A), which appear to correspond to the $CD8^-CD11b^{hi}$ and $CD8^+CD205^+$ lymphoid tissue DC subsets, respectively. CD11b expression on the $CD103^-$ DC population tends to be heterogeneous, and the $CD103^-CD11b^{low}$ subset may represent a less mature population. Corresponding counterparts to the $CD11b^{hi}$ and $CD103^+$ DC populations can be identified in human livers by the markers CD1c (BDCA1) and CD141 (BDCA3), respectively (Fig. 1B and C). The $CD1c^+$ population is somewhat heterogeneous for CD14 expression, suggesting that the $CD1c^{low}CD14^{hi}$ DC population may be analogous to the $CD11b^{low}CD103^-$ subset in mice. However, while some studies have defined $CD14^+$ and $CD16^+$ cells in the liver as DCs [3,9], it can be challenging to unequivocally distinguish these cells from monocyte and macrophage populations that may also express high levels of these markers, particularly in the setting of inflammation.

Studies of these DC subsets in other tissues indicate that the $CD11b^+$ subset produces higher levels of most cytokines and chemokines and efficiently processes and presents MHCII-restricted antigens to $CD4^+$ T cells, whereas the $CD103^+$ subset is more efficient at cross-presenting MHCI-restricted antigens to $CD8^+$ T cells [10–12] (Table 1). Studies in humans have similarly shown that $CD141^+$ DCs are more efficient at cross-presenting antigens than $CD1c^+$ DCs [3,13]. However, studies of $CD141^+$ DCs in the skin suggest that this subset may also serve a tolerogenic function by producing high levels of IL-10 and inducing regulatory T cells [14]. This is notable because studies of the total cDC

Table 1
Functional heterogeneity of dendritic cell subsets. While studies have yet to comprehensively elucidate the functional differences of hepatic DC subsets, studies of analogous DC subsets in other tissues suggest distinct functional characteristics. Based on data compiled from [1,10,11,13,14,17–19].

Species	DC type	DC subset	Key functional characteristics
Mouse	pDC • $Lineage^-$ • $CD11c^{int}$ • $MHCII^{int}$ • $PDCA\text{-}1^+$ • Siglec H^+		• MHC Class II presentation ± • MHC Class I cross-presentation ± (but can be induced by TLR stimulation) • Highly responsive to TLR7/9 ligands • Secrete high levels of IFNα during viral infections; negative regulators: Bst2 and Siglec H • Induction of IL-10 secreting Tregs (in vitro) • Induction of oral tolerance (in vivo) and tolerance in vascularized transplants
	cDC • $Lineage^-$ • $PDCA\text{-}1^-$ • $CD11c^{high}$ • $CD11c^{high}$ • $MHCII^{high}$	$CD103^+$ DC (Batf3 dependent DC)	• MHC Class II presentation + • High capacity of MHC Class I cross-presentation to cytotoxic T cells • Express TLR3 and TLR11-12 • Phagocytose apoptotic cells
		$CD103^-$ DC (Batf3 independent DC)	• High capacity for MHC Class II presentation of exogenous antigen • MHC Class I cross-presentation + • Express most TLRs and in addition RIG-1 and MDA5 • Secretes pro-inflammatory cytokines (TNFα and IL-6) after TLR ligation • A subsets with high production of TNFα and iNOS (TIPS DC, monocyte-derived DC)
Human	pDC • $Lineage^-$ • $MHCII^{high}$ • $CD123^+$ • $BDCA\text{-}2^+$		• MHC Class II presentation ± • MHC Class I cross-presentation + • Highly responsive to TLR7/8/9 ligands • Secrete high levels of IFNα during viral infections; negative regulators: BDCA-2 and ILT-7
	cDC • $Lineage^-$ • $CD123^-$ • $BDCA\text{-}2^-$ • $MHCII^{high}$	$CD141^+$ cDC	• MHC Class II presentation + • MHC Class I cross-presentation ++ • Highly responsive to TLR3 ligands • Produce high levels of CXCL10, IL12p70, IFNβ IFNλ after TLR stimulation
		$CD1c^+$ cDC	• MHC Class II presentation + • MHC Class I cross-presentation + • Produce high levels IL-8 at baseline and IL-1β after TLR3 stimulation

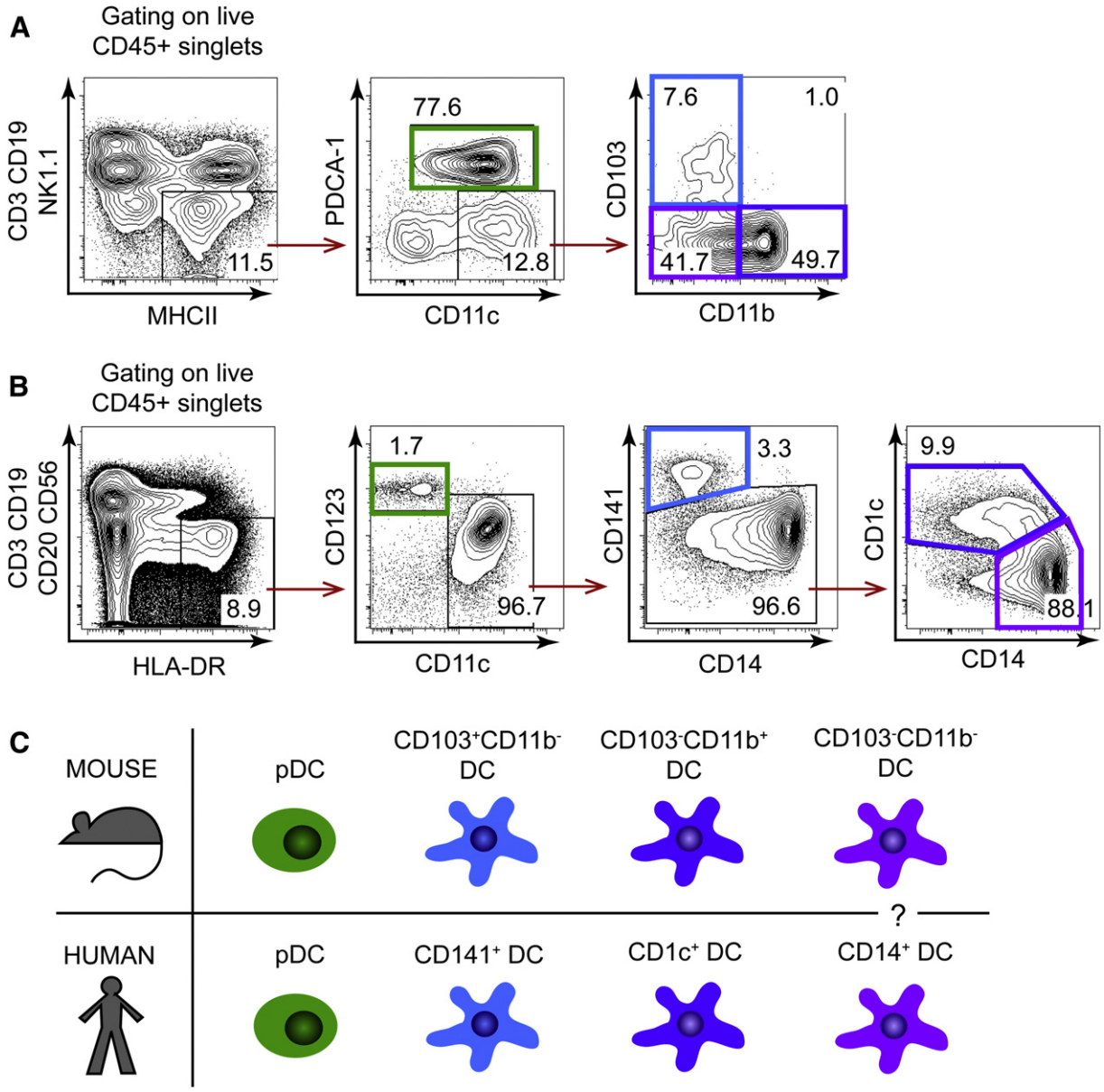

Fig. 1. Hepatic DC subsets in mice and humans. A liver sample from a naïve C57BL/6 mouse (A) and a liver resection sample from a colon cancer patient (B) were digested with 0.1 mg/mL collagenase IV and dissociated into single-cell suspensions. Liver mononuclear cells were isolated using a discontinuous 30%/70% percoll gradient, stained with fluorescently-labeled antibodies and analyzed by flow cytometry. In both mouse and human livers, DCs are contained with the population of cells expressing high levels of MHCII and lacking markers for other lineages. In mice, pDCs (green) can be identified based on high expression of PDCA-1. cDCs are contained within the $CD11c^{hi}$ population and can be further subdivided into $CD103^{+}CD11b^{low}$ (blue), $CD11b^{hi}CD103^{low}$ (dark purple) and $CD11b^{low}CD103^{low}$ (violet) populations. In human livers, pDCs (green) can be identified based on high expression of CD123. The cDC population contains a $CD141^{+}$ population (blue) that is believed to be analogous to the murine $CD103^{+}$ population and a $CD1c^{hi}$ population (dark purple) that is thought to be analogous to the murine $CD11b^{hi}$ population. The $CD14^{hi}CD1c^{low}$ population (violet) comprises another distinct population that may represent DCs but may also contain macrophages and monocytes. These populations are summarized in (C).

population in human livers indicate that, in comparison to DCs from the blood, spleen and skin, liver DCs produce higher levels of IL-10, which accounts for a reduced allogeneic stimulatory capacity [9,15]. Determining the relative cytokine profiles and T cell stimulatory capacities of the $CD1c^{+}$ and $CD141^{+}$ DC subsets in the liver remains an area of ongoing research.

Recent studies also suggest that the $CD141^{+}$ DC subset may play a specialized role during hepatic viral infections. The frequency of $CD141^{+}$ DCs is higher in the liver than in the peripheral blood and has been found to further increase in the setting of hepatitis C virus infection [16]. Studies of $CD141^{+}$ DCs in the blood indicate that this subset expresses high levels of TLR3 and responds to TLR3 ligands by producing high levels of CXCL10, TNFα and type III IFN [3,13,17]. Furthermore, recent studies suggest that while pDCs are the primary producers of type I IFN in response to HCV infection, $CD141^{+}$ DCs are the primary producers of type III IFN [18,19]. However, it should be noted that these findings have been established using blood-derived DCs, and whether CD141 DCs in the liver exhibit similar responses remains to be demonstrated.

3. Liver DC development and homeostasis

The heterogeneity and complexity of DCs has historically presented a challenge in accurately determining their ontogeny. While it is clear that tissue-resident DCs derive from hematopoietic stem cells in the bone marrow, the details of their developmental and differentiative pathways and the identities of their immediate precursors have only recently been elucidated and remain areas of active research. Early adoptive transfer experiments in mice demonstrated that DCs can develop from both the common lymphoid progenitor (CLP) and the common myeloid progenitor (CMP) in the bone

marrow, though the relative abundance of CMPs suggests that they are more likely to account for the majority of DCs in the periphery [20,21].

DC development is known to depend on FMS-like tyrosine kinase 3 (Flt3) ligand, and mice lacking Flt3 ligand expression show a dramatic reduction in the frequency and absolute number of DC populations in lymphoid and non-lymphoid tissues including the liver (Fig. 2). Flt3 (also known as fetal liver kinase-2, Flk2) expression has therefore been used to identify lineage-restricted precursors in the DC developmental pathway (Fig. 3). The CMP population is heterogeneous for Flt3 expression, and the $Flt3^+$ fraction gives rise to macrophage-DC progenitors (MDPs), which are more restricted precursors that are no longer able to differentiate into granulocytes, megakaryocytes or erythrocytes but can still produce monocytes, macrophages and DCs [22]. The MDPs give rise to common-DC progenitors (CDPs), which are able to produce cDCs and pDCs but not monocytes [23]. The CDP subsequently produces pDCs and pre-DCs, which exit the bone marrow and circulate through the blood and home to lymphoid and peripheral tissues and ultimately differentiating into mature DCs [24].

Adoptive transfer and genetic experiments indicate that the $CD103^+$ DC population in non-lymphoid tissues derives primarily from pre-DCs, while the $CD103^-$ population is more heterogeneous and may also derive from circulating monocytes [25–27]. As further evidence of this developmental heterogeneity, it is worth noting that there is a more profound reduction in the pDC and $CD103^+$ cDC subsets than in the $CD103^-$ subsets in the livers of $Flt3l^{-/-}$ mice (Fig. 2), supporting the notion that a Flt3-independent precursor contributes more appreciably to the $CD103^-$ DC population in the liver.

Recent studies of the transcription factor networks that govern DC development further reinforce the developmental heterogeneity amongst DC subsets. pDC development is specifically controlled by the E protein E2-2 and also requires interferon-regulatory factor 8 (IRF8) but is independent of inhibitor of DNA binding 2 (ID2) [28]. A number of studies have recently identified zDC (Zbtb46, Btbd4) as a transcriptional repressor that is specifically expressed by cDCs and distinguishes them from pDCs and monocytes [29–31]. Within the cDC population, $CD103^+$ non-lymphoid tissue DCs require IRF8, ID2 and basic leucine zipper transcriptional factor ATF-like 3 (BATF3), whereas $CD103^-$ DCs can develop independently of these factors [27,32].

Circulating monocytes likely contribute to some extent to the BATF3-independent DC population under steady state conditions. However, the contribution of monocytes to tissue DC populations changes dramatically under inflammatory conditions, when CCR2- and CX3CR1-expressing monocytes are recruited into tissues and differentiate into $CD11b^+$ monocyte-derived inflammatory DCs (moDCs) [33]. These infiltrating moDCs have been found to be important sources of TNFα and inducible nitric oxide synthase (iNOS) in the setting of bacterial and parasitic infection and may also contribute to pathological liver inflammation [34,35]. In mice, the circulating pre-DCs that give rise to the BATF3-dependent DC subset have been identified as a rare population of circulating $CD11c^+MHCII^-SIRP\alpha^{low}$ cells [24,36]; however, an analogous pre-DC population has yet to be identified in humans. It should be noted that human blood contains readily identifiable populations of $CD1c^+$, $CD141^+$ and $CD123^+$ DCs, which presents the possibility that these circulating DCs may contribute to the development of cDC and pDC populations in peripheral tissues.

Non-lymphoid tissue DCs continually sample antigens from the local microenvironment and DC activation in response to stimuli promotes their migration out of non-lymphoid organs through the afferent lymphatics (Fig. 3). The composition and homeostatic dynamics of liver DC populations can change considerably under inflammatory conditions, and the recruitment of other inflammatory cells into the liver further decreases their relative frequency. Consequently, the absolute number and relative proportions of the various DC populations in human liver resections and explants vary considerably with disease etiology and the extent of fibrosis (A.R. & C.A. unpublished observations). Given that both DC recruitment and migration are increased in the inflammatory state, the absolute number/frequency at a specific time-point may not entirely reflect the flux of DCs through a non-lymphoid organ. For a resident cell population (e.g., macrophages),

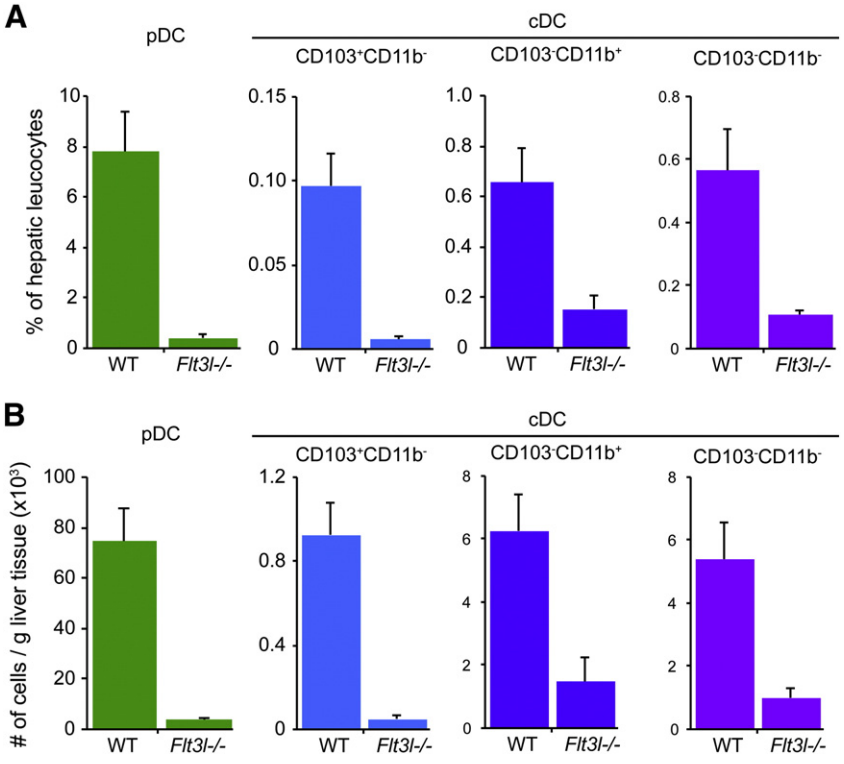

Fig. 2. Reduction in hepatic DC subsets in the absence of Flt3 ligand. Livers from WT and $Flt3l^{-/-}$ C57BL/6 mice were processed and analyzed as described in Fig. 1. The frequency (A) and absolute number (B) of the various DC populations was determined. Graphs indicate mean + SD of 4 mice per group.

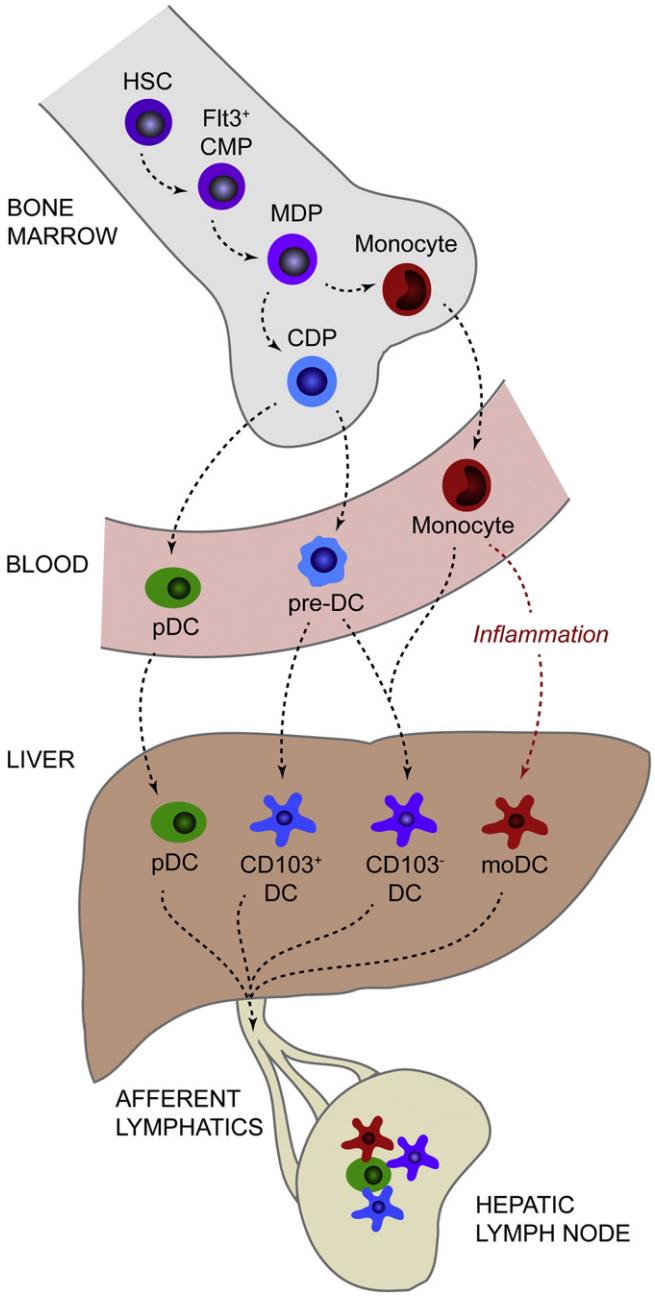

Fig. 3. Proposed model of liver DC development in mice. DCs develop from a Flt3$^+$ progenitor in the bone marrow. This progenitor gives rise to more restricted MDPs, which, in turn, give rise to CDPs and monocytes. CDPs give rise to pDCs and pre-DCs, which circulate through the blood and differentiate into liver DCs. The CD103$^+$ population is believed to arise almost exclusively from circulating pre-DCs, while the CD103$^-$ population is believed to derive partially from circulating monocytes. In the setting of inflammation, monocytes additionally contribute to hepatic DCs by differentiating into a distinct population of inflammatory monocyte-derived DCs. All of these DC subsets undergo homeostatic turnover in the liver and migrate to the draining hepatic lymph nodes through the afferent lymphatics.

the steady-state cell number is a reflection of recruitment (from progenitors or already mature cells), local proliferation and destruction (apoptosis, necrosis), whereas in the case of DCs one must also consider their kinetics of migration into and out of the organ.

4. Liver DC and fibrogenesis

Fibrosis is the liver's natural wound healing response to chronic injury, regardless of the etiology. A key feature of fibrosis is the activation of collagen producing cells including hepatic stellate cells, portal fibroblasts, bone marrow derived fibrocytes and fibroblast-like cells resulting from epithelial–mesenchymal transition. Additionally, hepatic exposure to injurious agents promotes robust immune system activation. Despite their low frequency within the total hepatic CD45$^+$ population, DCs may affect fibrosis through multiple potential mechanisms:

- DCs control the number and activity of many cells known to be critical in fibrogenesis thereby indirectly controlling fibrosis. For example, DCs have been shown to be critical in natural killer (NK) cell homeostasis and are the most potent cells involved in promoting CD8$^+$ T cell differentiation, both of which have been implicated in liver fibrosis.
- DCs can secrete significant levels of metalloproteinases (MMP) in both mice and humans, which appear to be critical to their migration out of the liver [37–39]. DC migration is the primary functional property that distinguishes DCs from macrophages and promotes the initiation of adaptive cellular immune responses in lymphoid organs. As discussed earlier, DCs are ontogenetically related to macrophages and at least a subset of CD11b$^+$ DCs originates from monocytes. Consequently, many functions classically attributed to the monocyte/macrophage lineage during tissue remodeling may in fact be DC dependent. Moreover, the studies using gadolinium chloride and liposomal clodronate that explored the role of macrophages/Kupffer cells on fibrosis in wide variety of models may also affect DC populations due to their high endocytotic activity and perivascular localization. In addition, both methods of treatment are non-specific and associated with bone marrow cell mobilizations [40], potentially including DC progenitors.

There are only a few studies that have explored the role of DCs in fibrogenic models of liver injury and some major limitations of these studies must be taken into account:

- The majority of the studies focused on the effect of DCs on liver inflammation as the end point during fibrogenesis rather than their effect on fibrosis.
- The immunophenotype of DCs was frequent limited to CD11c$^+$ cells and did not take into account the promiscuity of the CD11c$^+$ marker.
- Modulation of DC populations during fibrogenesis/fibrolysis has been accomplished by using transient approaches such as conditional depletion of CD11c$^+$ cells in transgenic mice expressing the human diphtheria toxin receptor (CD11c-DTR mice) and syngeneic transfers of purified DC. New insights into DC biology have led to the development of transgenic mice with constitutively impaired DC subsets, and these fibrogenesis/fibrolysis experiments have not been verified in these new strains of mice.
- Each of these studies only employed a single model of hepatic fibrosis (cholestatic or toxin-induced) and did not validate their findings using a second model.

To the best of our knowledge, there are no published reports that explore the effects of DCs on fibrogenesis. However, a possible involvement of DCs in fibrosis progression is suggested by two publications that evaluated the inflammation present in murine models of chronic liver injury and fibrosis. In the first report, Bleier et al. assessed the kinetics and function of liver DCs in the bile-duct ligation (BDL) model of liver fibrosis [41]. The authors are the first to explore the dynamics of DC populations after BDL and report a significant expansion of hepatic DCs with an enhanced capacity for stimulating allogeneic and syngeneic T cell response. While there was minimal ex vivo production of inflammatory cytokines, they found increased production of IL-6 following lipopolysaccharide exposure. DC recruitment was mediated by Gr1$^+$ cells and implicates a possible role for monocyte chemotactic protein 1 (MCP-1), which is known to promote DC recruitment and expansion. DCs were identified after NK exclusion as a CD11b$^+$CD11c$^+$ population, though their identity was not confirmed

by morphological and functional (DC migration) criteria. Furthermore, a significant proportion of the $CD11b^{+}CD11c^{+}$ cells in this study expressed low levels of MHCII, suggesting a significant contamination with other $CD11c^{+}$ cells (e.g., monocytes). In spite of these caveats, their data strongly support the concept of monocyte-derived DC recruitment in the BDL model of liver fibrosis.

Using a leptin/thiocetamide model of liver fibrosis [42], Connolly et al. also report significant recruitment of $CD11c^{+}$ cells into the liver with an enhanced capacity for antigen presentation and increased ex vivo and cytokine production with and without toll-like receptor (TLR) stimulation [43]. Their results also suggest a major role for DCs in influencing the hepatic cytokine microenvironment: diphtheria toxin (DT) induced-depletion of $CD11c^{+}$ cells in this model resulted in a significant decrease in tumor necrosis factor alpha (TNFα) production by leukocytes isolated from injured livers. Furthermore, coculture experiments indicated that these DCs enhance proliferation responses and cytokine production by hepatic stellate cells, suggesting a mechanism by which they may contribute to fibrosis progression. It should be noted however, that this study identified hepatic DCs by CD11c expression alone without excluding other cells that may express this marker in the setting of inflammation (NK cells and $CD11c^{+}$ monocytes), and the frequency of DCs within the liver was as high as 25% of the total hepatic leukocyte population, which is surprising given the typically scarcity of DCs in non-lymphoid tissues. The relatively low MHCII expression on the "$CD11c^{+}$ DC" suggests that this population may contain a significant number of other $CD11c^{+}$ cells (e.g., monocytes). Furthermore, some of the findings in this model may not be applicable to other models of liver fibrosis, given that leptin is well established to have direct effects on DC development, maturation and survival [44–46].

Our group has focused on the potential role of DCs during hepatic fibrosis regression [47]. Using multi-color flow cytometry we have meticulously identified the hepatic DC population by excluding lineage positive cells and have confirmed the identity of our DCs with morphological analysis after cytospin. Moreover, we have shown that the population up-regulates the lymph node homing receptor CCR7 and co-stimulatory molecules after TLR stimulation. Depleting DCs using a CD11c-DTR transgenic mouse model resulted in delayed fibrosis regression, though the use of only a single model for murine DC depletion may somewhat limit the interpretation of our data. The presence of neutrophilia after DC depletion in CD11c-DTR mice has recently been reported in murine infectious models [48–50]. In our studies we did not explore whether functional antagonistic properties of neutrophils (the deleterious effects of reactive oxygen species on the one hand and the capacity to secrete metalloproteinases that facilitate fibrosis resolution on the other) were affected by DC depletion and responsible for our observations. Consistent with our findings with DC depletion, parallel experiments inducing DC expansion by Flt3 ligand or adoptive cell transfer resulted in an acceleration of early fibrosis regression in an MMP-9-dependent mechanism and was, surprisingly, NK cell-independent. Ideally, these findings still need to be confirmed in a MMP-9 deficient DC mouse model. These findings suggest that DC expansion may be an effective therapeutic strategy to promote fibrosis regression in patients after the etiologic fibrotic agents has been removed (e.g., the clearance of hepatitis C virus).

It should also be noted that the role of DCs in fibrosis modulation is not limited only to liver. DCs have been identified as critical regulators in bleomycin-induced skin fibrosis, renal fibrosis induced by ureteral obstruction and bleomycin-induced lung fibrosis [51].

In conclusion, DCs play a potential role in liver fibrosis through the regulation of other immune cells (e.g., NK, $CD8^{+}$ T cells, hepatic stellate cells) and by secreting metalloproteinases. The few published reports point to the importance of the DC population in fibrogenic liver injury models but the use of improved genetic models and the standardization of DC identification using multi-color flow cytometry and morphological characteristics are required.

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