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SHORT COMMUNICATION

In vivo conversion of BM plasmacytoid DC into CD11b$^+$ conventional DC during virus infection

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DC are a highly heterogeneous population that plays a critical role in host defense. We previously demonstrated that virus infection induces BM plasmacytoid DC (pDC) differentiation into CD11b$^+$ conventional DC (cDC) upon *in vitro* culture with Fms-like tyrosine kinase 3 ligand (Flt3L). Here we use immunoglobulin D-J rearrangements and pDC adoptive transfer to provide definitive proof supporting BM pDC conversion into CD11b$^+$ cDC during *in vivo* viral infection. We show that *in vivo* BM pDC conversion into CD11b$^+$ cDC relates to enhanced ability to prime virus-specific T cells. Furthermore, we demonstrate that *in vivo* pDC conversion does not rely on viral infection of BM pDC, but instead is mediated by type I IFN signaling. Finally, by exploiting recently identified pDC-specific Ab, we provide further characterizations of the BM pDC fraction that exhibits this broader developmental plasticity. Collectively, these data indicate that BM pDC actively contribute to the CD11b$^+$ cDC pool during *in vivo* viral infection and delineates molecular, functional, and phenotypic features of this novel developmental pathway.

Key words: CD11b$^+$ conventional DC · Plasmacytoid DC · Type I interferon · Virus infection

Supporting Information available online

Introduction

DC play a central role in innate and adaptive immunity [1–3] and are composed by a large variety of subsets differing in tissue locations, surface phenotypes, and immunological functions [4, 5]. In mouse spleen, three major DC subsets have been identified including CD11b$^+$ DC (originally named myeloid DC), CD8α^+ DC (originally named lymphoid DC), and plasmacytoid DC (pDC) [6–8]. Typically, CD11b$^+$ DC and CD8α^+ DC are categorized as conventional DC (cDC) that are set apart from pDC in phenotypic and functional features. pDC, a unique DC subset with plasma cell-like morphology defined as CD11c$^+$B220$^+$ CD11b$^-$ in mice, produce large quantities of type I IFN (IFN-I) upon viral infection and represent a crucial element in antiviral immunity (reviewed in [9–11]).

The development of distinct DC subsets and their lineage relationship comprise a complex process that could be influenced by microenvironment changes such as pathogen infection or inflammation (reviewed in [12]). To gain more insight into the impact of virus infection on DC differentiation and the distribution of DC precursors, we used lymphocytic choriomeningitis virus (LCMV) infection in its natural host, the mouse, as a model system. In an earlier study, we demonstrated that BM pDC isolated from LCMV-infected mice can convert into CD11b$^+$ cDC

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in vitro [13]. Herein we provide direct evidence that conversion of BM pDC into CD11b$^+$ cDC is an ongoing process during *in vivo* viral infection. Moreover, we show that the reprogramming of pDC *in vivo* is independent of direct infection of pDC, but instead requires IFN-I signaling. Finally, we describe a CD11c$^+$B220$^+$ CD11b$^-$CD3$^-$NK1.1$^-$CD19$^-$Siglec-H$^-$ BM fraction (hereafter referred to as Siglec-H$^-$ pDC) that increases during LCMV infection in an IFN-I-dependent manner and exhibits high capacity to generate CD11b$^+$ cDC. Altogether, our studies reveal that the BM pDC serve as an alternative source to generate CD11b$^+$ cDC during *in vivo* viral infection and this process is mainly mediated by IFN-I signaling.

Results and discussion

BM pDC convert into CD11b$^+$ cDC during *in vivo* viral infection

Earlier we showed that BM, but not spleen, pDC isolated from LCMV-infected mice can differentiate into CD11b$^+$ cDC *in vitro* in the presence of Flt3L [13]. The same phenomenon was observed when challenging mice with poly(I:C) [13] and murine cytomegalovirus (MCMV) (data not shown), indicating that BM pDC conversion is a general event after virus infection. However, the essential biological question raised from this finding was whether the differentiation of pDC into CD11b$^+$ cDC occurs *in vivo*. Given that the phenotypic and functional properties of pDC have been changed after conversion, we used an intrinsic permanent marker, the D-J rearrangements of the IgH, as an indicator to monitor pDC-derived CD11b$^+$ cDC *in vivo* [14, 15]. For this, we isolated splenic CD11b$^+$ cDC from uninfected and LCMV Clone 13 (Cl 13) infected mice and the IgH D-J rearrangements were analyzed by a PCR-based approach (Fig. 1A). Importantly, the purity of sorted CD11b$^+$ cDC was over 98% and the percentage of contaminating B cells, T cells ,or NK cells was less than 1% (Supporting Information Fig. 1A). As expected, large amounts of D-J rearrangements were detected in B cells but no visible signal could be observed in granulocytes. In line with the findings by others [14, 15], the rearrangements of IgH were detected in splenic pDC but not in CD11b$^+$ cDC isolated from naïve mice. Remarkably, a significant increase in D-J rearrangements were detected in CD11b$^+$ cDC from LCMV Cl 13 infected mice, while the V-DJ rearrangements, a feature of B-cell lineage, were

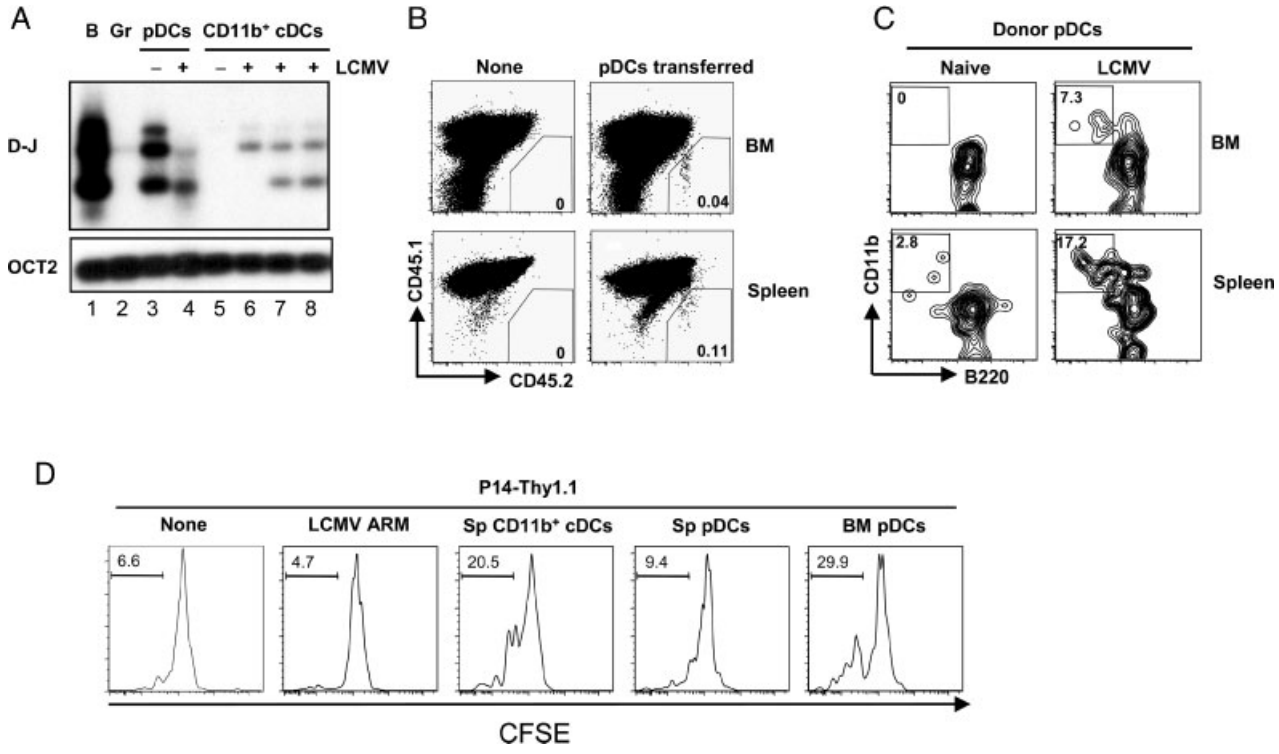

Figure 1. Reprogramming pDC into CD11b$^+$ cDC *in vivo* after LCMV infection. (A) FACS-purified splenic B cells, granulocytes, pDC, and CD11b$^+$ cDC from naïve or LCMV Cl 13 infected mice (at 3 dpi) were processed for PCR-based detection of IgH rearrangements. OCT-2 was measured as internal control. Lanes 6–8 show isolated CD11b$^+$ cDC from three individual experiments. (B and C) Adoptive transference of FACS-purified BM pDC from uninfected or LCMV-infected mice (CD45.2) into congenic mice (CD45.1). Numbers in each region indicate the frequency of CD45.2$^+$ donor cells (B) and the frequency of CD11c$^+$CD11b$^+$B220$^-$ cDC derived from donor pDC (C) in BM and spleen of recipient mice as indicated. (D) A total of 2×10^6 CFSE labeled Thy1.1$^+$D^{b}GP$_{33-41}$ specific CD8$^+$ T cells (P14-Thy1.1) were transferred into MHC class I D$^{b-/-}$ mice. The following day, recipient mice were either untreated, infected with LCMV ARM, or transferred with sorted spleen CD11b$^+$ cDC, spleen, or BM pDC from LCMV Cl 13 infected mice as indicated. Nine days after DC were transferred, CFSE dilution of P14-Thy1.1 was analyzed.

undetectable (Fig. 1A and Supporting Information Fig. 1B, respectively). Although indubitably detected, the intensity of the IgH D-J rearrangements in the $CD11b^+$ cDC was lower than the bands observed in pDC and B cells. This could result from the intrinsic heterogeneity of the $CD11b^+$ cDC population during infection. Indeed, other $CD11b^+$ cDC precursors, such as monocytes, have been demonstrated to contribute to the $CD11b^+$ cDC pool during inflammation [16]. Thus, pDC-derived-$CD11b^+$ cDC containing IgH D-J rearrangements likely represent only a fraction of the $CD11b^+$ cDC generated during the infection, in agreement with the lower intensity of the IgH D-J signal in this cell population. Altogether, these data suggest that a significant proportion of $CD11b^+$ cDC is derived from pDC *in vivo* after viral infection.

To further prove the reprogramming of pDC *in vivo*, we transferred FACS-purified BM pDC from naïve or LCMV Cl 13 infected C57BL/6 mice (CD45.2) into congenic (CD45.1) mice. Four days after transfer, BM and spleens from recipient mice were obtained and the donor cells ($CD45.2^+$) were identified by flow cytometry (Fig. 1B). Given that a fraction of BM pDC are infected with LCMV Cl 13, the recipient mice become infected upon cell transfer (data not shown). When transferring pDC from naïve mice, none or marginal levels of $CD11b^+$ cDC derived from donor pDC were detected in BM and spleen (Fig. 1C). In contrast, donor pDC from LCMV Cl 13 infected mice gave rise to approximately 15–20% of $CD11b^+$ cDC in spleen and an ~7% in BM. These $CD11b^+$ cDC did not result from contamination with BM proliferating progenitors as indicated by minimal cell division after cell transfer (Supporting Information Fig. 2). Collectively, these results reveal that BM pDC actively differentiate into $CD11b^+$ cDC during *in vivo* viral infection and provide new insights into the interrelationship of different DC subsets upon microbial invasion. Our data suggest that the addition of pDC-derived $CD11b^+$ cDC increases the heterogeneity of the cDC pool during viral infection.

pDC conversion relates to enhanced antigen presenting capacity *in vivo*

To gain more insight into the phenotypic and functional properties of *in vivo* generated pDC-derived $CD11b^+$ cDC, we transferred BM pDC into congenic recipients as described in Fig. 1B. We first investigated the morphology and expression of antigen-presenting molecules on pDC-derived $CD11b^+$ cDC in comparison with their counterparts existent in the recipient mice (Supporting Information Fig. 3). We found that the pDC-derived $CD11b^+$ cDC exhibit similar morphology, expression of CD11c, costimulatory molecule B7.2 and MHC class II molecules as the $CD11b^+$ cDC from recipient origin.

To evaluate the ability of pDC-derived $CD11b^+$ cDC to prime LCMV-specific T cells *in vivo*, we utilized a system in which $Thy1.1^+$, TCR transgenic CD8 T cells (P14-Thy1.1) specific for the LCMV $GP_{33–41}$ epitope presented in the context of H-$2D^b$ were labeled with CFSE and transferred into C57BL/6 $D^{b-/-}$ ($Thy1.2^+$) mice. Twenty-four hours later, DC from LCMV infected WT mice, were transferred into the same C57BL/6 $D^{b-/-}$ mice. The extent of CFSE dilution (*i.e.* proliferation) of P14-Thy1.1 cells was analyzed by FACS at day 9 post-DC-transfer (Fig. 1D). In this system the donor DC, expressing H-$2D^b$ molecules, are the only cells that could prime LCMV-specific-P14 T cells, as indicated by the lack of P14 division when $D^{b-/-}$ mice were infected with LCMV without receiving WT-DC. Transfer of BM pDC from LCMV infected mice (which differentiate into $CD11b^+$ cDC *in vivo*; Fig. 1C) induced significant proliferation of P14-Thy1.1 cells to a similar extent as spleen $CD11b^+$ cDC from LCMV-infected mice. In contrast, transfer of spleen pDC from the same LCMV-infected mice (which do not generate $CD11b^+$ cDC; [13]) induces background levels of T-cell proliferation. These data indicate that in the described experimental conditions pDC themselves are poor APC and are limited in priming virus-specific T cells during LCMV infection. Therefore, the T-cell expansion observed in $Db^{-/-}$ mice transferred with BM-pDC is most likely triggered by the pDC-derived-$CD11b^+$ cDC generated *in vivo* after cell transfer. These findings support the idea that $CD11b^+$ cDC derived from BM pDC can prime virus-specific T cells during LCMV infection to similar extents as endogenous $CD11b^+$ cDC existent in the spleen. These data are in agreement with our previous *in vitro* observations that pDC-derived $CD11b^+$ cDC are *bona fide* cDC and emphasize the role of pDC conversion in enhancing antigen presentation during viral infection *in vivo*.

BM pDC conversion is dependent on IFN-I and does not require virus infection of pDC

We next asked whether direct viral infection of pDC was required for their differentiation into $CD11b^+$ cDC and if there was any association between pDC conversion and the ability of the virus to establish chronic infection. To this end, we exploited the fact that while LCMV Cl 13 robustly replicates in DC and persists in most tissues ~60 days post-infection (dpi), its parental strain Armstrong 53b (ARM) exhibits minimal DC infection and is cleared within 7–10 dpi [17]. We hypothesized that if infection of pDC was required for this differentiation process, BM pDC from ARM-infected mice should be unable to generate $CD11b^+$ cDC. On the contrary, we found that pDC isolated from both ARM- and Cl 13-infected mice gave rise to a significant amount of $CD11b^+$ cDC after culture with Flt3L for 4 days (Fig. 2A). Moreover, when we analyzed the expression of LCMV nucleoprotein (NP) in cultures of BM pDC from Cl 13-infected mice, we observed that less than 10% of pDC-derived $CD11b^+$ cDC express LCMV NP *versus* 25–30% of the remaining pDC (Fig. 2B), indicating that infected pDC do not exhibit preferential conversion into $CD11b^+$ cDC over uninfected pDC in the same culture. Finally, we evaluated the presence of pDC-derived-$CD11b^+$ cDC *in vivo* during ARM and Cl 13 infection by examining IgH rearrangements in splenic $CD11b^+$ cDC at different time points post-infection. In accordance with our *in vitro* results, we detected a substantial amount of IgH rearrangements in $CD11b^+$ cDC from both LCMV ARM- and Cl 13-infected mice (Fig. 2C), suggesting

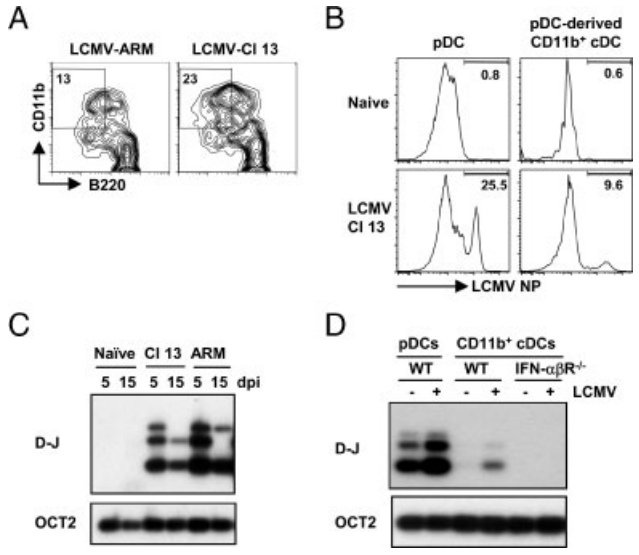

Figure 2. Conversion of pDC into CD11b$^+$ cDC is independent of pDC infection. (A) FACS-purified BM pDC from LCMV ARM or Cl 13-infected mice at 3 dpi were cultured with Flt3L for 4 days. The frequency of resulting CD11b$^+$ cDC is shown. (B) BM pDC were sorted from uninfected and LCMV Cl 13 infected mice and cultured as in (A). LCMV-NP in pDC and pDC-derived CD11b$^+$ cDC was detected by FACS and the frequency of NP-expressing cells is indicated. (C) FACS-purified splenic CD11b$^+$ cDC from uninfected, LCMV ARM, and Cl 13 infected mice at the indicated times p.i. were processed for PCR-based detection of IgH D-J rearrangements. A representative result from 2–3 independent experiments is shown. (D) WT and IFN-αβR$^{-/-}$ mice were infected with LCMV Cl 13 for 5 days. IgH D-J rearrangements in FACS-purified splenic pDC and CD11b$^+$ cDC were detected by PCR-based method. Results are representative of at least three independent experiments.

that the differentiation of pDC into CD11b$^+$ cDC *in vivo* occurs during infection with both acute (ARM) and persistent (Cl 13) viruses.

We next sought to address whether IFN-I was involved during the *in vivo* conversion of pDC into CD11b$^+$ cDC. To this end, we analyzed IgH rearrangements in splenic CD11b$^+$ cDC from WT and IFN-α and IFN-β-receptor-deficient (IFN-αβR$^{-/-}$) mice either uninfected or infected with LCMV. While significant D-J rearrangements were detected in splenic CD11b$^+$ cDC isolated from LCMV-infected WT mice, only background levels were found in their counterparts from IFN-αβR$^{-/-}$ mice (Fig. 2D), indicating that the lack of IFN-I signaling hampers the reprogramming of BM pDC *in vivo*.

Taken together, these data indicate that reprogramming of BM pDC during *in vivo* viral infection does not rely on direct infection of pDC and is instead the IFN-I signal that determines their differentiation into CD11b$^+$ cDC.

Differential capacity of BM pDC subtypes to give rise to CD11b$^+$ cDC

Recently, several molecules have been identified as pDC-specific markers including Ly49Q [18, 19], BM stromal cell antigen 2 (BST2, mAb 120G8, and PDCA-1) [20], and Siglec-H (mAb 440c) [21]. Among them, Ly49Q was defined to divide BM pDC into two sub-populations [18, 21]. We next investigated whether these two pDC subtypes have differential capability to convert into CD11b$^+$ cDC.

When we stained Ly49Q on BM pDC, we observed similar proportions of Ly49Q$^+$ and Ly49Q$^-$ cells in BM from uninfected and LCMV-infected mice. Interestingly, after 4 days of culture with Flt3L, Ly49Q$^+$ pDC, as well as Ly49Q$^-$ pDC gave rise to similar proportions of pDC-derived CD11b$^+$ cDC (Fig. 3A), indicating that both Ly49Q$^+$ and Ly49Q$^-$ pDC possess similar plasticity to differentiate into CD11b$^+$ cDC regardless of their distinct differentiation states. Our results differ from a recent report showing that Ly49Q$^+$ and Ly49Q$^-$ BM pDC isolated from poly(I:C) treated BALB/c mice have differential capacity to reprogram into CD11b$^+$ cDC [22]. This dissimilarity could be due to variations between stimuli, mouse strains, or technical differences.

Siglec-H plays an inhibitory effect on IFN-I production from activated pDC under cross-linking conditions [21, 23]. When we used the 440c mAb to detect Siglec-H, over 98% of BM pDC from uninfected mice were Siglec-H$^+$ (Fig. 3B). Notably, after LCMV infection, 20–30% of BM pDC were Siglec-H$^-$. BM stromal cell antigen 2 (BST2) antigen was expressed at comparably high levels in both Siglec-H$^+$ and Siglec-H$^-$ pDC (Supporting Information Fig. 4A). Moreover, even when BST2 was also up-regulated in other BM leukocytes after LCMV infection, their mean fluorescence intensity was much lower compared with that of pDC from infected mice (Supporting Information Fig. 4B and C). We further sorted Siglec-H$^+$ and Siglec-H$^-$ BM pDC from LCMV-infected mice and cultured them in the presence of Flt3L. Interestingly, the Siglec-H$^-$ pDC from LCMV-infected mice gave rise to a great proportion of CD11b$^+$ cDC, whereas Siglec-H$^+$ pDC generated only ~6% of CD11b$^+$ cDC (Fig. 3B). It should be noted that the reduced conversion in the Siglec-H$^+$ pDC cultures could be related to the inhibitory properties of the 440c Ab previously described [21, 23] and not necessarily indicate lack of potential to generate CD11b$^+$ cDC. Interestingly, Siglec-H$^-$ pDC isolated from LCMV-infected mice derived into Siglec-H$^+$ pDC, as well as into CD11b$^+$ cDC, after culture in the presence of Flt3L (Fig. 3C), suggesting that Siglec H$^-$ pDC could be either an earlier DC precursor or the resultant of internal redistribution of the heterogeneous BM pDC. Since the Siglec-H$^-$ pDC subset that exhibit high developmental plasticity is detected only after viral infection and the reprogramming of pDC is mediated by IFN-I, we hypothesized that IFN-I could be critical to generate the Siglec-H$^-$ pDC subset, which can subsequently differentiate into CD11b$^+$ cDC. To test this hypothesis, we infected IFN-αβR$^{-/-}$ mice with LCMV Cl 13 and analyzed the expression of Siglec-H in BM pDC at 3 dpi. Remarkably, only a minimal percentage of Siglec-H$^-$ pDC could be detected in BM from infected IFN-αβR$^{-/-}$ mice compared with the WT controls (Fig. 3D). Together with Fig. 2D, these results suggest that one of the mechanisms by which IFN-I promotes pDC conversion is the induction of Siglec-H$^-$ pDC, which exhibit high plasticity to differentiate into CD11b$^+$ cDC.

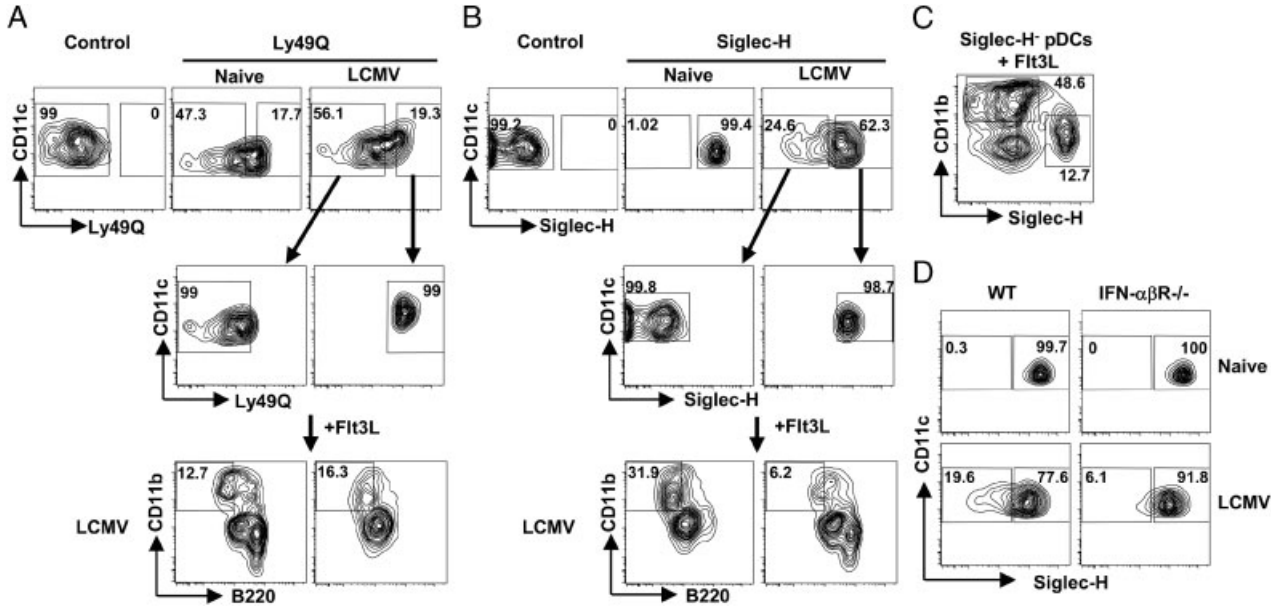

Figure 3. Siglec-H$^-$ pDC exhibit high plasticity to differentiate into CD11b$^+$ cDC. BM pDC were isolated from uninfected or LCMV Cl 13-infected mice and further divided into two subpopulations based on their expression of Ly49Q (A) or Siglec-H (B). Upper panels, pre-sort gating for Ly49Q and Siglec-H staining. Middle panels, purity of FACS-purified populations from LCMV-infected mice. Lower panels, frequency of pDC-derived CD11b$^+$ cDC 4 days after Flt3L-culture. (C) Siglec-H$^-$ pDC from LCMV Cl 13 infected mice were isolated and cultured for 4 days in the presence of Flt3L. Numbers within regions indicate the frequency of CD11b$^+$ cDC and Siglec-H$^+$ DC derived in the culture. (D) Siglec-H expression on BM pDC from WT and IFN-αβR$^{-/-}$ mice at day 3 p.i. with LCMV Cl 13. Each representative data set was obtained from at least three independent experiments.

Concluding remarks

With the notion that CD11b$^+$ cDC serve as a better APC to prime T cells [24, 25], the reprogramming of pDC into CD11b$^+$ cDC may result in the increase in APC and thus facilitate the switch from innate into adaptive immunity. Indeed, our data support the idea that while pDC themselves are limited at priming antiviral T cells, pDC-derived-CD11b$^+$ cDC contribute to antigen presentation and expansion of virus-specific T cells during LCMV infection. Previous studies have demonstrated that pDC are potent producers of IFN-I during LCMV infection [26, 27]. Therefore, pDC conversion may have evolved as a mean to switch from a poor antigen-presenting interferon producing DC subset (pDC) to a more potent antigen-presenting DC subpopulation (CD11b$^+$ cDC). The host would benefit from this cell conversion by maximizing antigen presentation and T-cell priming to fight the infection while preventing sustained IFN-I production that could cause immunopathology. On the other hand, re-direction of BM pDC into CD11b$^+$ cDC may result in reduced pDC numbers and subsequently compromise the production of IFN-I after secondary infections. Finally, it is possible that pDC-derived CD11b$^+$ cDC would play a unique role in anti-viral defense different from other CD11b$^+$ cDC (*e.g.* monocyte-derived-CD11b$^+$ cDC) that account for their emergence upon virus infection.

Our present work demonstrates that BM pDC conversion into CD11b$^+$ cDC is an ongoing process during *in vivo* viral infection and better characterizes the mechanisms, the populations involved, and biological significance of this cellular event. This novel pathway of DC differentiation is turned on during viral infection and actively contributes to the ensuing DC pool to fight the infection. These findings further highlight the flexibility and the dynamics of the DC subsets in response to the micro-environmental changes, which may be critical to mount the most appropriate type of immune response against the invading pathogens.

Materials and methods

Viruses and Ab

LCMV Armstrong clone 53b and Cl 13 stock were generated, genotyped, and quantified by plaque assay as described previously [28]. The following Ab were purchased from either eBioscience (San Diego, CA) or BD Biosciences (San Jose, CA): anti-CD3, anti-CD19, anti-Ter119, anti-Ly6G (clone 1/A8), PE or APC anti-CD11c, PE or PerCp-Cy5.5 anti-CD3, CD19, and NK1.1, PE-Cy7 anti-CD11b, PE anti-B7.2 (CD86), Alexa-700 anti-MHC class II, APC-Cy7 anti-B220, Pacific Blue anti-CD8, and APC streptavidin. Anti-120G8 mAb was kindly provided by Dr. Giorgio Trinchieri (National Institutes of Health, Frederick, USA) and conjugated with Alexa-488 (Molecular Probes) in our laboratory. Biotin-conjugated Siglec-H Ab clone 440c was provided by Dr. Marco Colonna. Biotin-conjugated Ly49Q Ab was obtained from

Dr. Noriko Toyama-Sorimachi (Tokyo Medical and Dental University Graduate School, Tokyo, Japan). Anti LCMV-NP 113 mAb was produced in our laboratory.

Mice

C57BL/6 (CD45.2, Thy1.2$^+$), C57BL/6 CD45.1, C57BL/6 Thy1.1$^+$ D^bGP_{33-41} TCR transgenic, C57BL/6 IFN$\alpha\beta R^{-/-}$, and C57BL/6 $D^{b-/-}$ mice were obtained from the Rodent Breeding Colony at The Scripps Research Institute (TSRI). All mice were handled according to the NIH and TSRI Animal Research Guidance. Mice were infected with 2×10^6 pfu of LCMV ARM or Cl 13 i.v.

Cell isolation, flow cytometry, and FACS purification

BM and spleen cell isolation, flow cytometry, and FACS-purification were performed and analyzed as described previously [18]. pDC were defined as CD11c$^+$B220$^+$120G8^{high}-CD11b$^-$CD3$^-$NK1.1$^-$CD19$^-$ cells and CD11b$^+$ cDC as CD11c$^+$ CD11b$^+$B220$^-$CD3$^-$NK1.1$^-$CD19$^-$ cells. Cell purity was exceeded 98%.

Adoptive transference

To transfer pDC into congenic mice, sorted pDC (1×10^6 cells) from C57B6-CD45.2 mice were i.v. injected into congenic C57BL/6-CD45.1 female mice after a sub-lethal irradiation dose (600 rad). Four days after transfer, CD11c$^+$ cells were enriched from BM or spleen with CD11c MACS beads according to the manufacturer's instructions (Miltenyi Biotec, Auburn, CA). Positive selected cells were incubated with fluorochrome-conjugated-specific Ab and analyzed by flow cytometry (LSRII, BD Biosciences).

Rearrangements of immunoglobulin heavy chain

The preparation of DNA and the PCR assays for detecting D-J and V-DJ rearrangements of the immunoglobulin heavy chain (IgH) were described previously [29, 30]. It is important to note that all PCR including the control OCT-2 have been tested in the linear phase of amplification. Therefore, the OCT-2 signal represents the equal amount of input DNA for PCR.

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Conflict of interest: The authors declare no financial or commercial conflict of interest.

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Abbreviations: **ARM:** Armstrong 53b · **BST2:** BM stromal cell antigen 2 · **cDC:** conventional DC · **Cl 13:** clone 13 · **dpi:** days post-infection **IFN-I:** type I IFN · **LCMV:** lymphocytic choriomeningitis virus · **NP:** nucleoprotein · **pDC:** plasmacytoid DC · **TSRI:** The Scripps Research Institute

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