Expansion of polymorphonuclear myeloid-derived suppressor cells in patients with end-stage renal disease may lead to infectious complications

Yan-Fang Xing[1,2,8], Rui-Ming Cai[3,8], Qu Lin[4,8], Qing-Jian Ye[5], Jian-Hua Ren[6], Liang-Hong Yin[2] and Xing Li[4,7]

[1]Department of Nephrology, the Third Affiliated Hospital of Guangzhou Medical University, Guangzhou, People's Republic of China; [2]The First Affiliated Hospital of Jinan University, Guangzhou, People's Republic of China; [3]Department of Urology, the Third Affiliated Hospital of Guangzhou Medical University, Guangzhou, People's Republic of China; [4]Department of Medical Oncology, the Third Affiliated Hospital of Sun Yat-sen University, Guangzhou, People's Republic of China; [5]Department of Gynecology, the Third Affiliated Hospital of Sun Yat-sen University, Guangzhou, People's Republic of China; [6]Department of Orthopedics, the Third Affiliated Hospital of Sun Yat-sen University, Guangzhou, People's Republic of China; and [7]Guangdong Key Laboratory of Liver Disease Research, the Third Affiliated Hospital of Sun Yat-sen University, Guangzhou, People's Republic of China

Myeloid-derived suppressor cells (MDSCs) are recently identified immune suppressive cells in multiple chronic inflammations. Here, we investigated MDSCs in patients with end-stage renal disease (ESRD) and their clinical significance in these patients and healthy individuals (49 each). Polymorphonuclear and mononuclear MDSCs were investigated by flow cytometry. Patients with ESRD before hemodialysis presented a significantly higher level of polymorphonuclear MDSCs. Depletion of polymorphonuclear-MDSCs resolved T cell IFN-γ responses. By co-culture, T cell proliferation and the production of IFN-γ were abrogated by the addition of polymorphonuclear MDSCs in a dose-dependent manner. Both of these effects were reversed by a reactive oxygen species inhibitor. The levels of reactive oxygen species were higher in polymorphonuclear MDSCs derived from patients with ESRD than from normal individuals. The mRNA level of NOX2, the key protein complex responsible for reactive oxygen species production, was higher in ESRD-related polymorphonuclear MDSCs. The phospho-STAT3 level, a key activator of MDSCs, was higher in ESRD-related polymorphonuclear MDSCs. Finally, the polymorphonuclear MDSC level before and after hemodialysis was positively related to infectious diseases. Patients with ESRD were dichotomized into 2 groups by the amount of polymorphonuclear MDSCs. Patients with high levels of polymorphonuclear MDSCs presented with a higher incidence of infectious events. Thus, polymorphonuclear MDSCs were elevated in ESRD patients with strong immune-suppressive capability through a phospho-STAT3/reactive oxygen species pathway. Hence, polymorphonuclear MDSCs might increase the risk of infectious complications.

Kidney International (2017) **91,** 1236–1242; http://dx.doi.org/10.1016/j.kint.2016.12.015

KEYWORDS: end-stage renal disease; myeloid derived suppressor cell; reactive oxygen species

Copyright © 2016, International Society of Nephrology. Published by Elsevier Inc. All rights reserved.

Infectious diseases are the major causes of death for end-stage renal disease (ESRD) patients with AIDS as 1 of the critical causes.[1,2] Chronic kidney disease (CKD) results in accumulation of low–molecular mass metabolites, including phenylacetic acid, homocysteine, various sulfates, guanidine compounds, and others, which have inhibitory effects on immune cell activation, promote leukocyte apoptosis, and induce the oxidative burst in phagocytes.[3,4] Recently, novel mechanisms have come to light, including elevation of Treg cells, activation of T cells, and neutrophil responsiveness to a bacterial challenge.[5,6] However, few therapies have been developed to counteract the immune deficiency of patients with ESRD, partially due to lack of practical targets.

Myeloid-derived suppressor cells (MDSCs) are recently identified immune suppressive cells with the ability to suppress T cell activation and function.[7,8] MDSCs were investigated initially in malignant diseases.[9] In recent years, it has become clear that MDSCs also play an important role in the regulation of immune responses in chronic inflammations.[10–12] ESRD entails chronic inflammation.[3] However, the role of MDSCs in ESRD has not been illustrated. Moreover, a series of targeted therapies have been identified to be effective in eliminating MDSCs,[9,13] which made MDSC a potentially practical target for immune therapy in ESRD patients.

MDSCs are now divided into 2 major populations: granulocytic or polymorphonuclear MDSC (PMN-MDSC) and mononuclear MDSC (M-MDSC).[7,9,14] In the present study, we investigated the 2 subtypes of MDSC in patients with ESRD and further illustrated their clinical significance.

Correspondence: *Xing Li, Department of Medical Oncology, the Third Affiliated Hospital of Sun Yat-sen University, 600 Tianhe Road, Guangzhou 510630, People's Republic of China. E-mail: lixing9@mail.sysu.edu.cn*

[8]These authors contributed equally to this work.

Received 7 September 2016; revised 1 December 2016; accepted 15 December 2016; published online 16 February 2017

RESULTS

PMN-MDSCs expansion in patients with ESRD

During the period between October 2015 and September 2016, we investigated a series of 49 patients with ERSD before initiation of dialysis in the Third Affiliated Hospital of Guangzhou Medical University and the Third Affiliated Hospital of Sun Yat-sen University, Guangzhou, China (Supplementary Table S1). The ESRD was considered irreversible by 2 independent nephrologists, rather than representing acute kidney injury.[6] Age- and gender-matched healthy controls ($n = 49$) consisted of local volunteers. Blood samples were collected at diagnosis of ESRD and at the pre-dialysis time points under the maintenance hemodialysis phase after a median of 8-month hemodialysis (HD) vintage. Peripheral blood mononuclear cells (PBMCs) were isolated from whole blood by Ficoll centrifugation and analyzed within 6 hours of blood sampling. Circulating frequencies of the subsets of MDSCs were quantified with the gating strategy indicated. Both the PMN-MDSCs and M-MDSCs were CD33 positive (Figure 1a). Patients with ESRD before dialysis presented with a significantly higher level of PMN-MDSCs, whereas the frequency of the M-MDSCs was not higher than that in healthy controls. Interestingly, after HD, the level of PMN-MDSCs decreased significantly (Figure 1b).

PMN-MDSCs suppress T cell activation

MDSCs are characterized by their suppressive capability on T cell response because they share the same definitive markers as their normal counterparts, PMN-MDSCs and M-MDSCs.[7] In order to investigate whether PMN-MDSCs and M-MDSCs in patients with ESRD suppress T cell response, PMN-MDSCs and M-MDSCs were removed by flow sorting, then PBMCs and $PBMC^{\Delta PMN\text{-}MDSC/M\text{-}MDSC}$ were stimulated with anti-CD3 and anti-CD28 for 4 days. The expression of interferon-γ (IFN-γ) by CD4 and CD8 T cells was tested to evaluate T cell response.[15] As expected, depletion of PMN-MDSC and M-MDSC did not change T cell response in healthy donor–derived PBMCs (Figure 2a and b), nor did depletion of M-MDSCs in patients with ESRD (Figure 2c). T cell IFN-γ responses were rescued by depletion of PMN-MDSCs (Figure 2d).

In order to confirm the immune suppressive capacity of PMN-MDSCs in patients with ESRD, T cells and PMN-MDSCs were purified from PBMC using flow sorting. Carboxyfluorescein diacetate succinimidyl ester (CFSE)–labeled PBMC-derived CD3+ T cells were stimulated with anti-CD3 and anti-CD28, with the indicated ratio of PMN-MDSCs. CD4+ and CD8+ T cell proliferations were almost completely abrogated by the addition of ESRD-related PMN-MDSCs at a 2:1 ratio. The addition of ESRD-related PMN-MDSCs resulted in significantly reduced proliferation of both CD4+ and CD8+ T cells in a dose-dependent manner. The IFN-γ levels in the media were tested using enzyme-linked immunosorbent assay (ELISA), which determined that IFN-γ secretion decreased after administration of ESRD-related PMN-MDSCs (Figure 3). The same cells from healthy donors did not exhibit a suppressive function, indicating that PMN-MDSCs exist in patients with ESRD rather than in healthy donors.

P-STAT3/ROS pathway is the mechanism for ESRD-related PMN-MDSC–mediated immune suppression

Based on the finding that PMN-MDSCs from patients with ESRD suppressed antigen-nonspecific T cell proliferation, we further explored the underlying mechanisms controlling PMN-MDSC–mediated T cell suppression. Previous reports had confirmed that production of arginase I or reactive oxygen species (ROS) were immune mediators for PMN-MDSC–mediated immune suppression.[8,14] Thus, we utilized the arginase inhibitor N(omega)-hydroxy-nor-l-arginine

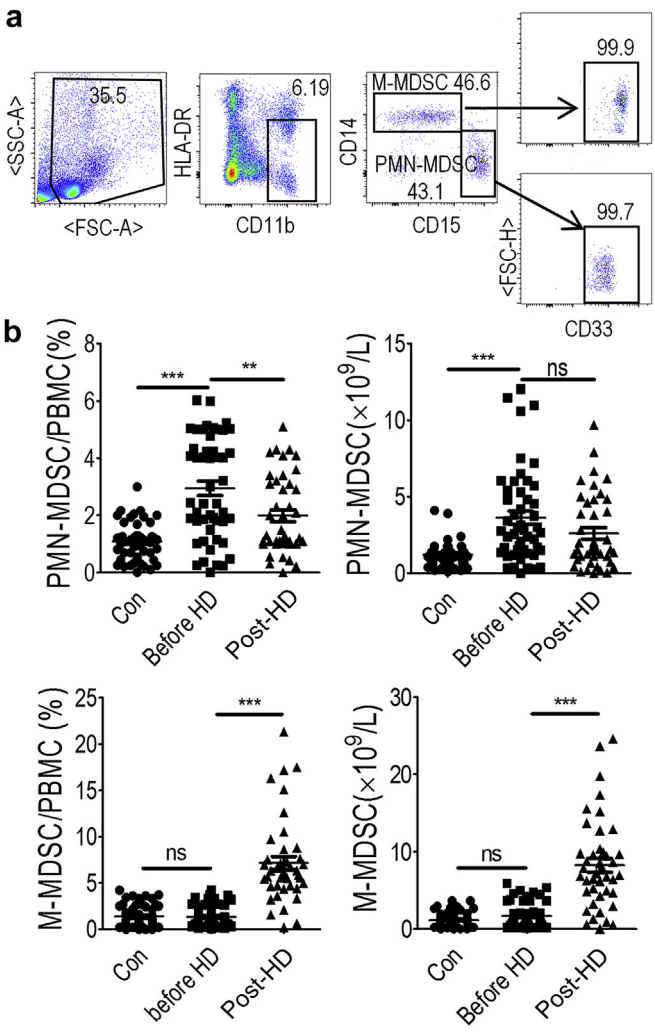

Figure 1 | Expansion of polymorphonuclear (PMN) myeloid-derived suppressor cells (MDSCs) dampens T cell function in patients with end-stage renal disease (ESRD). (**a**) Gating strategy of mononuclear MDSCs (M-MDSCs) by flow cytometry analysis. PMN-MDSC was defined as $CD11b^{+}CD15^{+}CD14^{-}HLA\text{-}DR^{-/low}$, with M-MDSC defined as $CD11b^{+}CD15^{-}CD14^{+}HLA\text{-}DR^{-/low}$. CD33 expression was evaluated among PMN-MDSCs and M-MDSCs. (**b**) Statistical analysis of PMN-MDSC and M-MDSC frequency in the peripheral blood of healthy controls and patients with ESRD before and after hemodialysis (HD). $^{**}P < 0.01$; $^{***}P < 0.001$.

Download English Version:

https://daneshyari.com/en/article/5688505

Download Persian Version:

https://daneshyari.com/article/5688505

Daneshyari.com