ORIGINAL ARTICLE

Basic Study

Role of cytomegalovirus on the maturation and function of monocyte derived dendritic cells of liver transplant patients

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Abstract

AIM: To study the impact of association between cytomegalovirus (CMV) pathogenesis with dendritic cell (DC) maturation and function was evaluated in CMV reactivated liver transplanted patients in comparing with non-reactivated ones, and healthy controls.

METHODS: Monocyte derived dendritic cells (MoDCs) was generated from collected ethylenediaminetetraacetic acid-treated blood samples from patient groups and controls. In these groups, expression rates and mean fluorescent intensity of DC markers were evaluated using flowcytometry technique. Secretion of cytokines including: interleukin (IL)-6, IL-12 and IL-23 were determined using enzyme-linked immunosorbent assay methods. The gene expression of toll-like receptor 2 (TLR2), TLR4 and IL-23 were analyzed using in-house real-time polymerase chain reaction protocols.

RESULTS: Results have been shown significant decreases in: Expression rates of MoDC markers including CD83, CD1a and human leukocyte antigen DR (HLA-DR), the mean fluorescence intensitys for CD1a and HLA-DR, and secretion of IL-12 in CMV reactivated compared

with non-reactivated liver transplanted patients. On the other hand, significant increases have been shown in the secretions of IL-6 and IL-23 and gene expression levels of TLR2, TLR4 and IL-23 from MoDCs in CMV reactivated compared with non-reactivated liver transplanted recipients.

CONCLUSION: DC functional defects in CMV reactivated recipients, such as decrease in expression of DC maturation markers, increase in secretion of proinflammatory cytokines, and TLRs can emphasize on the importance of CMV infectivity in development of liver rejection in transplanted patients.

Key words: Cytomegalovirus; Dendritic cells; Liver transplantation

Core tip: Cytomegalovirus (CMV) can interfere with maturation and antigen-presenting function of dendritic cell (DC). This interference with DC function could promote viral spread by paralyzing the adaptive immune system. CMV with DC infection induces inflammatory cytokines and activation of the interferon pathway in transplanted patients. DCs undergo lytic viral cycles, can induce late gene expression of CMV, release of infectious virus, and stimulating of T-cell responses resulted to allograft rejection.

INTRODUCTION

Liver transplantation is the definitive treatment of choice for patients with end-stage liver disease[1]. Graft rejection and infection remains major complications post liver transplantation[2]. In liver recipients, cytomegalovirus (CMV) is the most determinative viral infectious pathogen cause of morbidity and mortality post-liver engraft and associate with diminished graft survival[3]. This ubiquitous viral infection in immuno-compromised transplant patients belongs to family *Herpesviridae*, subfamily *Betaherpesvirinae*, genus human herpes virus 5, species *Cytomegalovirus*[4]. CMV primary infection results in life-long residence of the virus in the host, and reactivates in immuno-compromised individuals frequently. Reactivation of CMV infection and development of related severe diseases and syndromes are common in solid organ recipients may lead to severe complications following transplant, such as acute rejection[5,6]. CMV may lead also to higher

rates of bacterial and fungal infections in transplant recipients[7]. In transplant patients, CMV infection causes both direct effects, reflecting cell destruction and indirect effects, such as acute or chronic rejection[8]. Primary CMV infection induced immune related proinflammatory response that was maintained during latency. This continuous activation of the immune system may play a role in the acceleration of chronic diseases and pathogenesis of chronic allograft rejection[9,10].

CD14+ monocytes and/or myeloid progenitor cells are site of CMV latency and are capable of harboring quiescent viral genomes[11]. Monocyte represents a key cell type in the CMV pathogenesis, since mostly represent as an important cellular reservoir for latent virus[12,13]. A number of studies have shown that CMV infection in monocytes is non-permissive and cellular differentiation is prerequisite for CMV replication[11]. CMV replication can be reactivating in latently infected monocytes related to differentiation dependent manner[11]. The dendritic cells (DCs) generated from CMV infected monocytes. CMV infected monocyte derived DCs (CMV-MoDCs) have an altered phenotype and functional defects[14]. DCs are determinative initiators of cellular immunity against CMV infection[1]. DCs also act with superiority over other antigen-presenting cells (APCs) in stimulating T-lymphocyte responses and maintaining protective antiviral immunity[15].

CMV can interfere with maturation and antigen-presenting function of DCs and also disturb both innate and adaptive immunity[14,16]. This interference with DC function could promote viral spread by paralyzing the adaptive immune system[17]. CMV with DC infection induces many hallmarks of innate immunity, such as the production of inflammatory cytokines and activation of the interferon pathway in transplanted patients. This induction is rapid and can promote without requirement of CMV reactivation. DCs undergo lytic viral cycles, can induce late gene expression of CMV, release of infectious virus, and stimulating of T-cell responses resulted to allograft rejection[8].

Therefore, in this study the impact of association between CMV pathogenesis with DC maturation and function was evaluated in CMV reactivated liver transplant patients in comparing with non-reactivated ones and healthy controls.

MATERIALS AND METHODS

Patients and samples

Ten liver transplanted patients who admitted at Transplant Center of Namazi Hospital, Shiraz, Iran were enrolled in this study between years 2012 and 2014. These patients divided to two groups including: 5 patients with CMV reactivation and rest of them without CMV reactivation. Therefore, CMV reactivation was confirmed in these transplanted patients using antigenemia protocol. The CMV antigen positive cells were counted and positive results are reported as one or more CMV pp65 antigen infected cell per 50000

Table 1 Underlying diseases in liver transplanted patients	
Underlying diseases	**Patients (n = 10)**
PSC	2
Hypercholesterolemia	1
Cryptogenic cirrhosis	2
Hepatitis C virus infection	1
Hepatitis B virus infection	1
Wilson disease	1
NASH	1
Autoimmune hepatitis	1

PSC: Primary sclerosing cholangitis; NASH: Non-alcoholic steato-hepatitis.

Table 2 Average means fluorescence intensity for surface markers of monocyte derived dendritic cells in cytomegalovirus reactivated, non-reactivated liver transplanted recipients, and healthy individuals			
Surface marker	CMV reactivated patients	CMV non-reactivated patients	Healthy controls of MoDCs
	MFI ± SE	MFI ± SE	MFI ± SE
CD83	30.15 ± 1.06	32.3 ± 2.3	29.2 ± 2.5
CD86	100.5 ± 3.1	103 ± 4.5	83 ± 6.7
CD1a	47 ± 1	80 ± 3	53 ± 13
HLA-DR	49.3 ± 5.4	73.6 ± 6.5	55.8 ± 4.9

The data are the means ± SE. MFI: Mean fluorescence intensity; HLA-DR: Human leukocyte antigen DR; CMV: Cytomegalovirus; MoDC: Monocyte derived dendritic cell.

white blood cells (WBCs). In all 5 patients with CMV reactivation 5 to 7 CMV pp65 antigen infected cell was found per 50000 WBCs, with a mean of 6 ± 1.01 cells/50000 WBCs. Underlying diseases for studied liver transplanted patients have been shown in Tables 1 and 2. The 20 mL ethylenediaminetetraacetic acid (EDTA)-treated blood samples were collected from each evaluated transplant recipients.

Also, 20 mL EDTA-treated blood samples were collected from 5 healthy volunteers as healthy controls. The age range (20-50 years old) and male to female ratio were similar in studied transplanted patient groups and healthy controls. This study was in accordance with the ethical standards of Shiraz University of Medical Sciences Committee (the study protocol conformed to the ethical guidelines of the 1975 Declaration of Helsinki). The informed consent was obtained from each studied transplant patients. The immunosuppressive conditioning regimen used in liver transplanted patients was previously described[18].

CMV antigenemia assay
CMV antigenemia protocol was performed on the EDTA-treated blood samples to determine viral reactivation by evaluation of the presence of lower matrix pp65 antigen in polymorph nuclear cells using the CMV Brite Turbo kit (IQ Products, Groningen, Netherlands) according manufacturer instruction as previously described[19].

Generation of MoDCs
Leukocytes were isolated from 20 mL EDTA-treated blood samples collected from each liver transplanted patients with and without CMV reactivation using gradient centrifugation through Lymphodex (Inno-train, Germany), the cells from interphase were collected. CD14+ monocytes were isolated by positive selection using a MACS system (Miltenyi Biotech, Bergisch Gladbach, Germany), according to the manufacturer's protocol.

Monocytes were cultured in six-well cell culture plates in RPMI medium (Invitrogen, United States) supplemented with 4 mmol/L L-glutamine (Life technologies, United States), 100 IU/mL penicillin (Life technologies, United States), 10% heat-inactivated fetal bovine serum (Life technologies, United States), 1% sodium

pyruvate (Bioidea, Iran), 1% non-essential amino acid (Life technologies, United States), 1000 IU/mL recombinant human granulocyte macrophage-colony stimulating factor (R and D Systems, United Kingdom) and 500 IU/mL recombinant human interleukin-4 (IL-4; R and D Systems, United Kingdom) in a 37 ℃ 5% CO_2 humidified incubator. Every 3 d 200 μL of the medium was exchanged with fresh medium and cytokines. For mature cells, maturation was induced on day 5 by adding 1000 IU/mL recombinant human tumor necrosis factor-α (TNF-α; R and D Systems, United Kingdom) and allowing maturation to proceed for 48 h.

Analysis of MoDC markers
Matured MoDCs were harvested and stained with fluorescently labeled monoclonal antibodies including: PE-anti-CD14, FITC-anti-CD83, FITC-anti-CD86, FITC-anti-CD1a and FITC-anti-HLADR (eBiosciences, United States). Cell suspension was mixed with antibody solution followed by incubation in the dark at 4 ℃ for 45 min. Cells were suspended in phosphate buffer saline and data acquisition for 10000 events was performed using flowcytometry (Becton Dickinson, San Jose, United States). FlowJo software (Flexera Company, United States) was used to analyze the expression rate and mean fluorescence intensity (MFI) of studied DC markers.

Measurement of cytokine levels
The IL-6, IL-12 and IL-23 cytokines released from MoDCs in culture supernatant were measured using the commercial human enzyme-linked immunosorbent assay Ready-SET-Go kits (eBioscience, United States) according to the manufacturer's protocols.

The gene expression of cytokines and toll-like receptors
Total RNA was extracted from MoDCs using RNX plus (CinnaGen, Iran). RNA samples were reverse transcribed using Reverse Transcriptase (Vivantis, Malaysia) and random hexamer as previously described[20]. An amount of 1 μg total RNA was used to produce cDNA. The primers that were used to analyze the gene transcripts including: TLR-2 (NM_003264.3), TLR-4 (NM_003266.3),
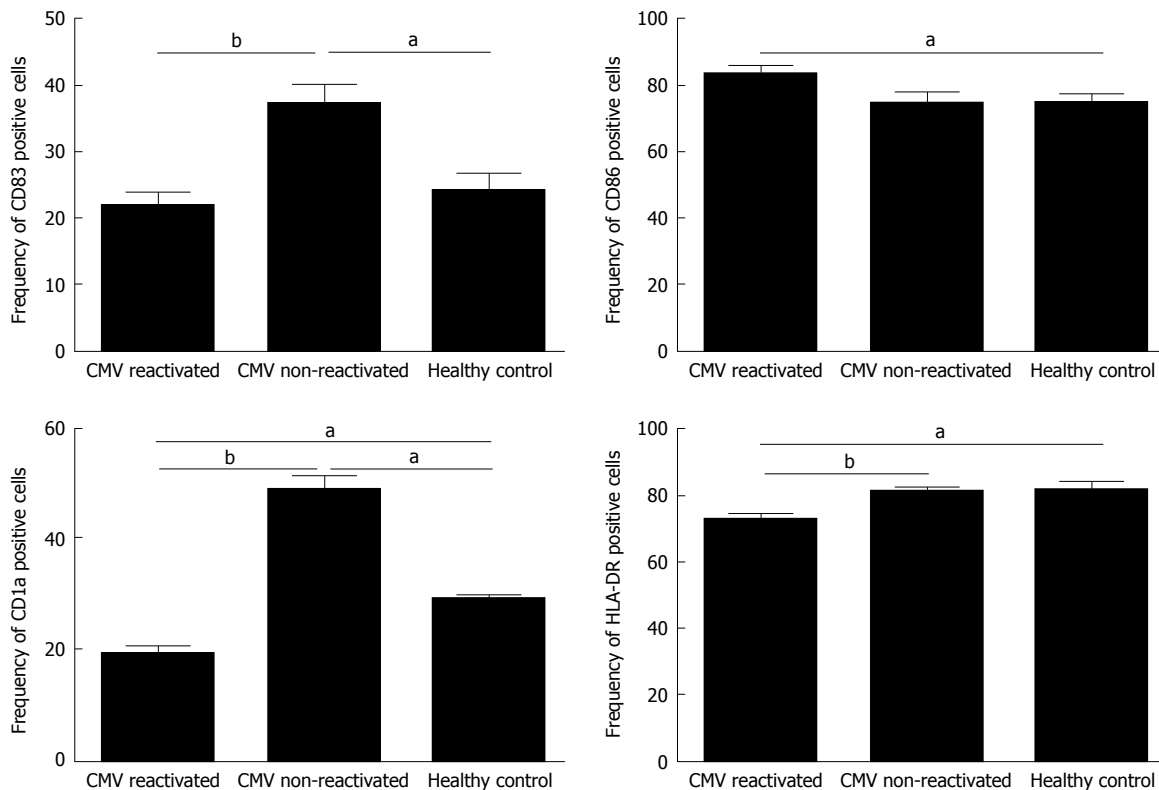
Figure 1 The expression rates for surface monocyte derived dendritic cell markers of CD83, CD86, CD1a and human leukocyte antigen DR in cytomegalovirus reactivated patients, cytomegalovirus non-reactivated recipients, and healthy control. The expression rates of CD83, CD1a and HLA-DR were significantly decreased in CMV reactivated patients *vs* non-reactivated recipients. Any significance is indicated [a]P < 0.05, [b]P < 0.01. The data are the means ± SE. HLA-DR: Human leukocyte antigen DR; CMV: Cytomegalovirus.

IL-23 (NM_016584.2) and β-actin (NM_001101.3)[18]. The mRNA expression levels of the IL-23, TLR2 and TLR4 genes were finally determined in MoDCs of liver transplanted patients with and without CMV reactivation compared with healthy controls using in-house-real time polymerase chain reaction (PCR) protocols as previously described[18].

The PCR reaction was carried out in a final volume of 20 μL containing: 10 μL SYBR green Premix by Ex taq (Takara, Japan), 0.4 μL SYBR Green Dye, 0.8 μL forward and 0.8 μL reverse primers (8 pmol), 6 μL H_2O and 2 μL cDNA template. The thermal cycling profile was the same for each primer set and consisted of an initial denaturation at 95 ℃ for 2 min, followed by 40 amplification cycles of 95 ℃ for 30 s and 65 ℃ for 20 s using Step One Plus Real-Time instrument (ABI, Step One Plus, United States). The mean Ct value of target genes in each sample was normalized using β-actin gene Ct value to give a ΔCt value. This was then normalized to healthy control (ΔΔCt), and finally the $2^{-\Delta\Delta Ct}$.

Statistical differences between studied groups were evaluated using non-parametric tests of version 15 of SPSS software (Chicago, United States). The sample analysis was also analyzed using version 5 of Graph Pad Prism software (United States). The $2^{-\Delta\Delta Ct}$ value was calculated using Livak method for analysis of the expression level of studied genes. The *P*-value of < 0.05 was considered as significant.

RESULTS

Expression of MoDC markers in patient groups and controls

The expression rate (*P* = 0.02) and MFI (*P* = 0.04) of CD86 were both significantly increased in CMV re-activated patients in comparing with healthy controls with median value of 82% *vs* 75%, respectively (Figures 1 and 2). The expression rate of CD1a was significantly decreased in CMV reactivated patients in comparing with healthy controls with median value of 18% *vs* 30%, respectively (*P* = 0.01) (Figure 3). The expression rate of human leukocyte antigen DR (HLA-DR) was significantly decreased in CMV reactivated patients in comparing with healthy controls with median value of 72% *vs* 84%, respectively (*P* = 0.01) (Figure 4).

The expression rate of CD83 was significantly increased in CMV non-reactivated patients in comparing with healthy controls with median value of 40% *vs* 21%, respectively (*P* = 0.02) (Figure 1). The MFI of CD86 was significantly higher in CMV non-reactivated patients than that in healthy control (*P* = 0.04) (Figure 2). The expression rate of CD1a was significantly raised in CMV non-reactivated patients than that in healthy controls with median value of 50% *vs* 30%, respectively (*P* = 0.01) (Figure 1). The MFI of HLA-DR was significantly raised in CMV non-reactivated liver transplanted patients than that in healthy controls (*P* =
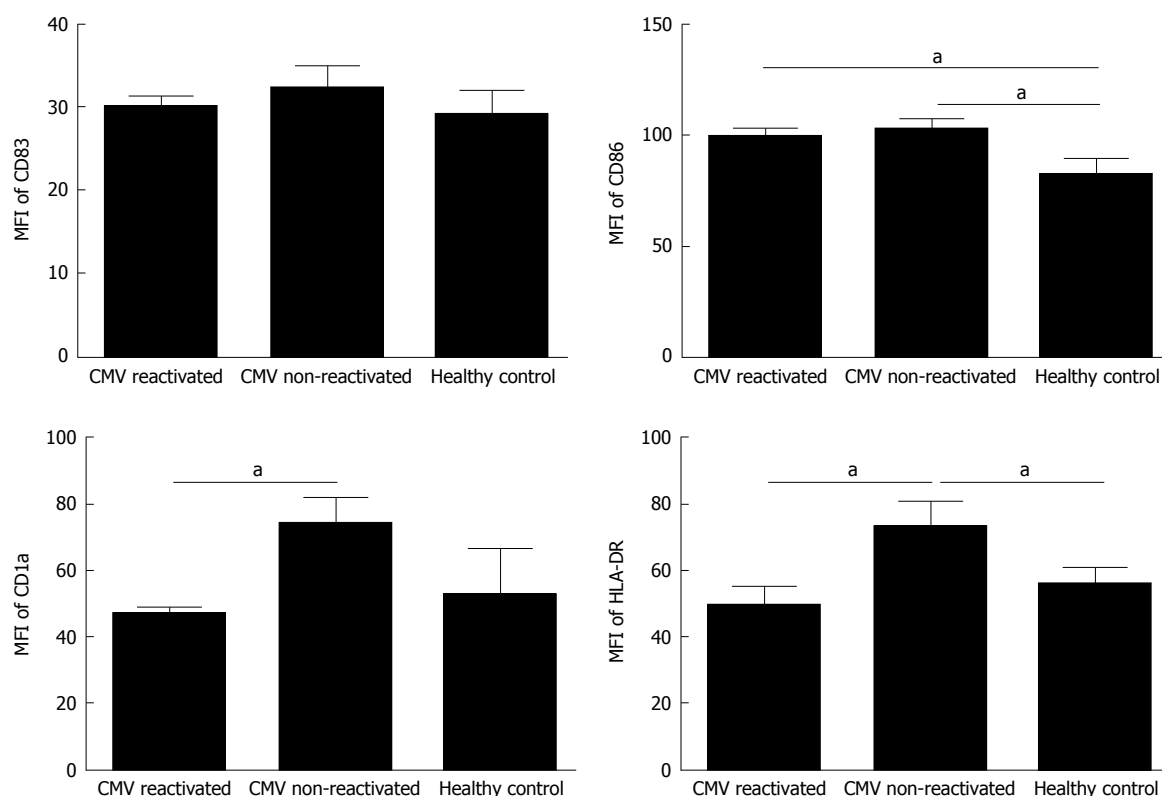
Figure 2 Mean fluorescence intensity for surface monocyte derived dendritic cell markers of CD83, CD86, CD1a and human leukocyte antigen DR in cytomegalovirus reactivated patients, cytomegalovirus non-reactivated recipients, and healthy control. The MFI of CD1a and HLA-DR were significantly decreased in CMV reactivated patients *vs* non-reactivated recipients. Any significance is indicated [a]$P < 0.05$. The data are the means ± SE. MFI: Mean fluorescence intensity; HLA-DR: Human leukocyte antigen DR; CMV: Cytomegalovirus.

0.03) (Figure 5).

Expression rate of CD83 was significantly decreased in CMV reactivated compared with non-reactivated liver transplanted patients with median value of 22% *vs* 40%, respectively ($P = 0.007$) (Figure 1). The expression rate ($P = 0.007$) and MFI ($P = 0.02$) of CD1a was significantly lower in CMV reactivated compared with non-reactivated patients with median value of 18% *vs* 50%, respectively (Figures 1 and 2). The expression rate ($P = 0.007$) and MFI ($P = 0.03$) of HLA-DR was significantly decreased in CMV reactivated patients compared with non-reactivated recipients with median value 72% *vs* 80%, respectively (Figures 1 and 2).

Cytokine secretions by MoDCs in patient groups and controls

The secretion of IL-6 from MoDCs of CMV reactivated patients was significantly increased in comparing with healthy control with mean of 334.6 ± 2.2 pg/mL *vs* 312 ± 1.08 pg/mL ($P = 0.009$) (Figure 6A). The secretion of IL-12 from MoDCs of CMV reactivated patients was significantly decreased in comparing with healthy control with mean of 1.26 ± 0.04 pg/mL *vs* 1.54 ± 0.03 pg/mL ($P = 0.01$) (Figure 6B). The IL-23 secretion level from MoDCs of CMV reactivated patients was significantly increased in comparing with healthy control with mean of 13.53 ± 0.09 pg/mL *vs* 13.1 ± 0.1 pg/mL ($P = 0.02$) (Figure 6C).

The secretion of IL-6 from MoDCs of CMV non-reactivated patients was significantly decreased in comparing with healthy control with mean of 261.2 ± 3.72 pg/mL *vs* 312 ± 1.08 pg/mL ($P = 0.006$) (Figure 6A). Also, secretion of IL-12 from MoDCs of CMV non-reactivated patients was significantly increased in comparing with healthy control with mean of 1.98 ± 0.03 pg/mL *vs* 1.54 ± 0.03 pg/mL ($P = 0.009$) (Figure 6B). The secretion of IL-23 from MoDCs of CMV non-reactivated patients was significantly decreased in comparing with healthy control with mean of 8.77 ± 0.19 pg/mL *vs* 13.1 ± 0.1 pg/mL ($P = 0.008$) (Figure 6C).

The secretion of IL-6 from MoDCs was significantly higher in CMV reactivated patients than that in CMV non-reactivated ones with mean of 334.6 ± 2.2 pg/mL *vs* 261.2 ± 3.72 pg/mL ($P = 0.005$) (Figure 6A). The secretion of IL-12 from MoDCs was significantly lower in CMV reactivated patients compared with non-reactivated ones with mean of 1.26 ± 0.04 pg/mL *vs* 1.98 ± 0.03 pg/mL ($P = 0.007$) (Figure 6B). The secretion of IL-23 from MoDCs was significantly higher in CMV reactivated patients than that in CMV non-reactivated ones with mean of 13.53 ± 0.09 pg/mL *vs* 8.77 ± 0.19 pg/mL ($P = 0.007$) (Figure 6C).

Cytokine and TLR gene expression by MoDCs in patient groups and controls

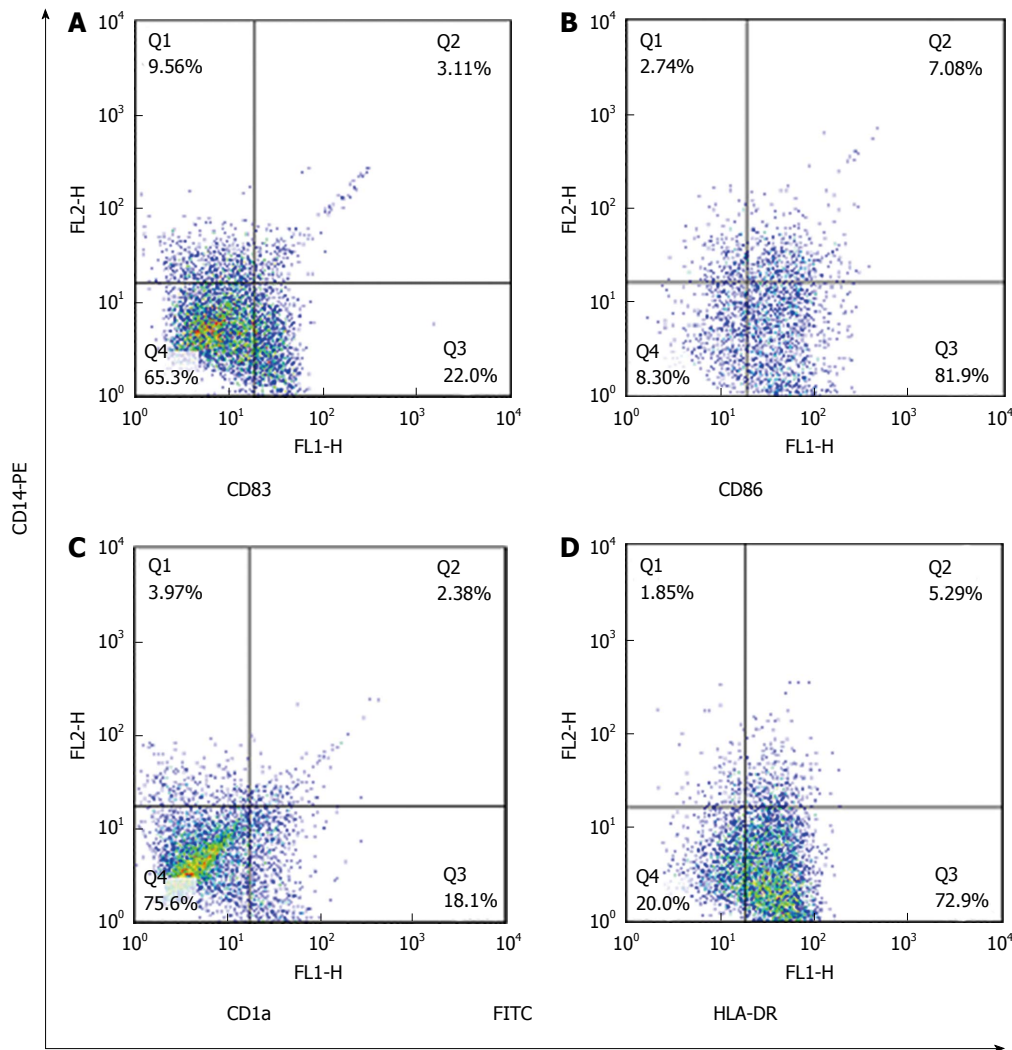The IL-23, TLR2, and TLR4 mRNAs was expressed 5.2

Figure 3 The expression rate of monocyte derived dendritic cell markers in cytomegalovirus reactivated patients was examined by dual-color cytometry. Expression of surface markers: CD83 (22%) (A), CD86 (82%) (B), CD1a (18%) (C) and HLA-DR (72.9%) (D) on MoDCs in CMV reactivated patients. CD14-PE, phycoerythrin-conjugated CD14, CD83-FITC, fluorescein isothiocyanate-conjugated CD83, CD86-FITC, fluorescein isothiocyanate-conjugated CD86, CD1a-FITC, fluorescein isothiocyanate-conjugated CD1a, HLADR-FITC, fluorescein isothiocyanate-conjugated HLA-DR. HLA-DR: Human leukocyte antigen DR; CMV: Cytomegalovirus; MoDC: Monocyte derived dendritic cell.

(P = 0.005), 3.6 (P = 0.007), and 4.3 (P = 0.009) folds significantly more in CMV reactivated patients compared with healthy controls, respectively (Figure 7).

The gene expression level of IL-23 was the same in both CMV non-reactivated patients and healthy controls (P = 0.6) (Figure 7A). The TLR2 mRNA expression was significantly decreased in CMV non-reactivated patients in comparing with healthy controls (P = 0.02) (Figure 7B). The IL-23, TLR2, and TLR4 mRNAs was expressed 4.7 (P = 0.007), 5.4 (P = 0.005), and 2.8 (P = 0.01) folds significantly more in CMV reactivated compared with non-reactivated patients, respectively (Figure 7).

DISCUSSION

DCs have superiority over other APCs in viral infections to stimulate T-cell responses and maintaining protective antiviral immunity[15]. DCs are critical initiators of cellular immunity against viruses especially CMV[1]. CMV

as a determinative human pathogen can cause fatal complications and promote rejection in transplanted recipients[21]. CMV infection can also increase the rate of immunosuppression with interfering the maturation and function of DCs post-transplantation[16,22]. Therefore, in this report, the effects of CMV reactivation compared to non-reactivation were elucidated on maturation and function of DCs in liver transplanted recipients and healthy controls.

CMV and DC maturation was interested of related researchers in earlier studies[16,22]. Down-regulation of major histocompatibility complex (MHC) class I and CD86 and CD83 costimulatory molecules on immature DCs and inhibition of DC maturation was indicated following *in vitro* CMV infection of DCs[16]. CMV targets DCs and alters their functions by interfering with MHC-II biosynthesis and maturation, as well as with the expression and function of related endocytic proteases[23]. CMV-infected DCs displayed abnormal
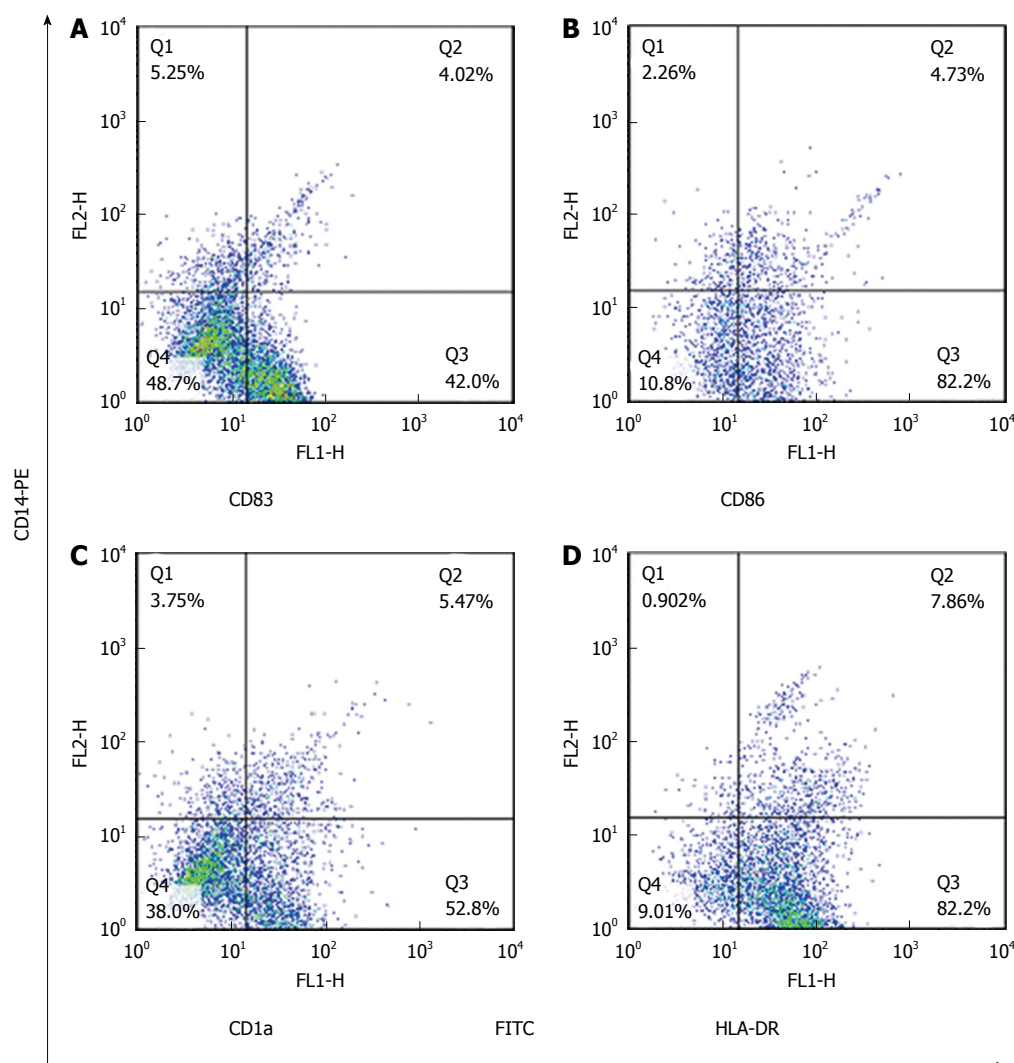
Figure 4 The expression rate of monocyte derived dendritic cells markers in cytomegalovirus non-reactivated patients was examined by dual-color cytometry. Expression of surface markers: CD83 (42%) (A), CD86 (82%) (B), CD1a (52.8%) (C) and HLA-DR (82%) (D) on MoDCs in CMV non-reactivated patients. HLA-DR: Human leukocyte antigen DR; CMV: Cytomegalovirus; MoDC: Monocyte derived dendritic cell.

phenotypic characteristic and stable expression of CD83 as a maturation marker[24,25]. Infectivity of CMV in MoDCs resulted in maturation of the surviving cells, up-regulation of the CD86, and down-regulation of MHC-Ⅰ and Ⅱ[21,26]. In this study, results also demonstrated the down-regulation of CD83, CD1a and HLA-DR molecules on MoDCs in CMV reactivated compared to non-reactivated liver transplanted patients. The MFIs of CD1a and HLA-DR were significantly down-regulated in CMV reactivated patients compared to non-reactivated ones. On the other hand CMV-mediated up-regulation of CD86 and down-regulation of CD1a and HLA-DR molecules were found in CMV reactivated patients compared to healthy controls. Therefore CMV interference with maturation of DCs promotes viral spread by paralyzing the adaptive immune system[17]. Especially, in transplanted recipients, CMV-infected DCs are less capable of developing antiviral activated APCs, this may lead to impaired immune responses not only against CMV, but most likely also against other invading microorganisms.

Stimulation of toll-like receptors (TLRs) on DCs activates signal transduction pathways lead to induction of a range of antimicrobial genes and inflammatory cytokines[11,27]. TLR signaling pathways trigger a series of interactions among specific intracellular mediators that ultimately result in the release of nuclear factor-κB (NF-κB) from its related endogenous inhibitors[11]. Earlier reports emphasized that markedly up-regulation of TLR2 and TLR4 responsible for early activation of alloimmune T-cells favoring to acute renal and also liver allograft rejection[28-30]. TLR2 was recently identified as a cell surface receptor activates secretion of inflammatory cytokine response to CMV infection[4,11]. *In vitro* stimulation of TLR2 by CMV resulted in NF-κB activation and cytokine secretion[31,32]. CMV was also able to activate TLR4 and mediate cytokine secretion in human monocytic cells[33]. Similarly, *TLR2* and *TLR4* gene expression by MoDCs was significantly increased in studied CMV reactivated liver transplanted patients compared to non-reactivated ones and healthy controls.

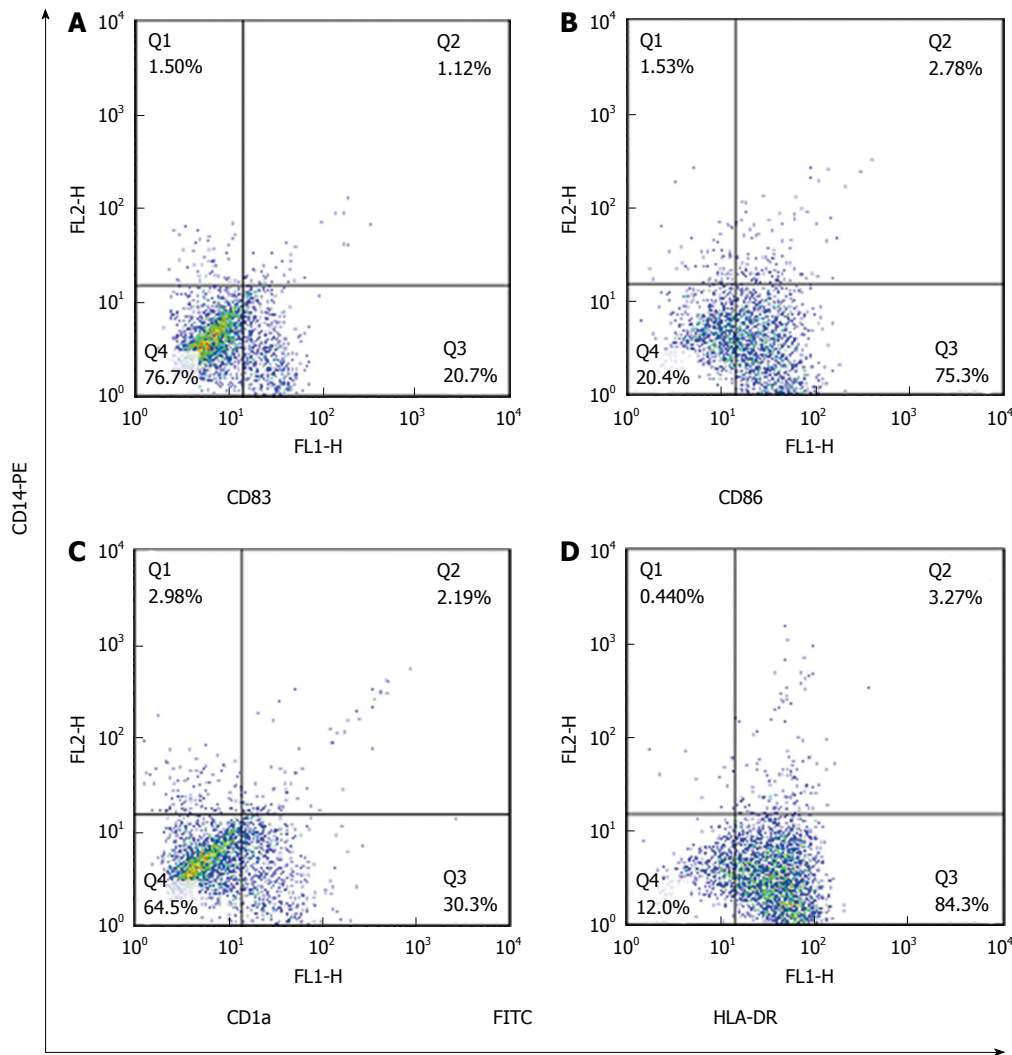Pathological processes associated with CMV reac-

Figure 5 The expression rate of monocyte derived dendritic cell markers in healthy control was examined by dual-color cytometry. Expression of surface markers: CD83 (20.7%) (A), CD86 (75.3%) (B), CD1a (30%) (C) and HLA-DR (84%) (D) on MoDCs in healthy control. HLA-DR: Human leukocyte antigen DR; MoDC: Monocyte derived dendritic cell.

tivation appear to be mediated by the release of inflammatory cytokines[11]. Following CMV infection DCs produce no IL-12 and only low levels of TNF-α[16]. Down-regulation of IL-12 production impairs the antiviral mechanisms of T cells and NK cells in patients with active CMV infection[34]. Similar to previous reports, results of the present study revealed that secretion of IL-12 by MoDCs was significantly decreased in CMV reactivated liver transplanted patients compared to CMV non-reactivated ones and healthy controls. But, Th17 cell linage and related cytokines like IL-17 and IL-23 have determinative role contribute to the mechanisms of allograft rejection[35-39]. IL-6 is also essential for differentiation of IL-17-producing human Th cells[40]. IL-6 and subsequent signaling pathways are important for activation and differentiating DCs. Activation and concomitant production of these cytokines also appear to be essential for reactivation and replication of CMV in infected patients such as transplant recipients[21]. Similarly, IL-6 and IL-23 secretion and expression by

MoDCs are significantly higher in CMV reactivated in comparing with non-reactivated patients and healthy controls.

In conclusion, results of this study highlight the fact that, CMV and DCs contractions promote different pathways including: Interference with the maturation and expression of DC markers (CD83, CD1a and HLA-DR), IL-12 decreasing and IL-6 and IL-23 elevation from MoDCs and also increase of the mRNA expression levels of *TLR2*, *TLR4* and *IL-23* genes in MoDCs of CMV reactivated liver transplanted patients. These pathways can implicate in the development of acute or chronic allograft liver rejection.
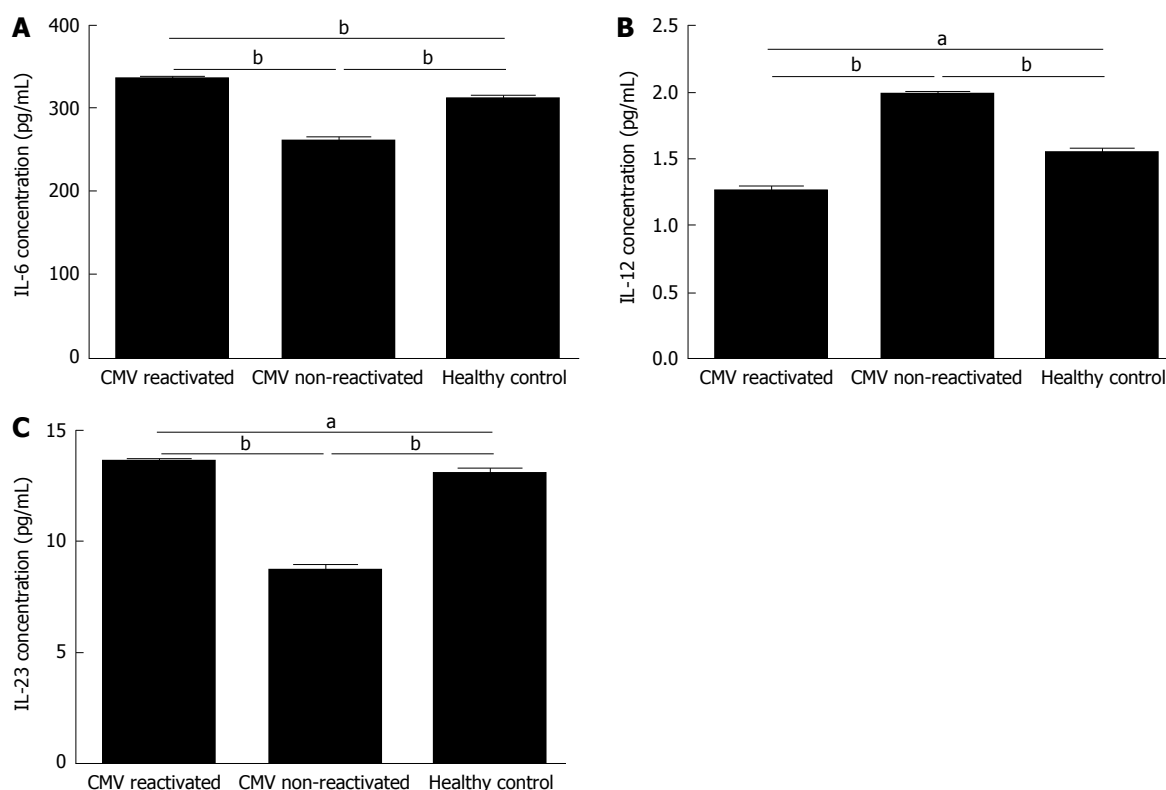
ACKNOWLEDGMENTS

Figure 6 Levels of cytokines secreted by monocyte derived dendritic cells in cytomegalovirus reactivated patients, non-reactivated recipients, and healthy controls. The IL-6, IL-12 and IL-23 concentrations in the supernatants were measured by enzyme-linked immunosorbent assay. IL-6 and IL-23 secretions were significantly increased in CMV reactivated patients compared to non-reactivated recipients and healthy controls (A and C). IL-12 secretion was significantly decreased in CMV reactivated patients compared to non-reactivated ones and healthy controls (B). Any significance is indicated [a]$P < 0.05$, [b]$P < 0.01$. The data are the means ± SE. CMV: Cytomegalovirus; IL: Interleukin.

Figure 7 In cytomegalovirus reactivated patients, non-reactivated recipients, and healthy controls, mRNA relative expressions of interleukin-23 (A), toll-like receptor 2 (B) and toll-like receptor 4 (C) were determined by real-time polymerase chain reaction protocols. The gene expression levels of IL-23, TLR2 and TLR4 were significantly increased in CMV reactivated patients in comparing with non-reactivated ones and healthy controls. Any significance is indicated [a]$P < 0.05$, [b]$P < 0.01$. The data are the means ± SE. CMV: Cytomegalovirus; IL: Interleukin; TLR: Toll-like receptor.
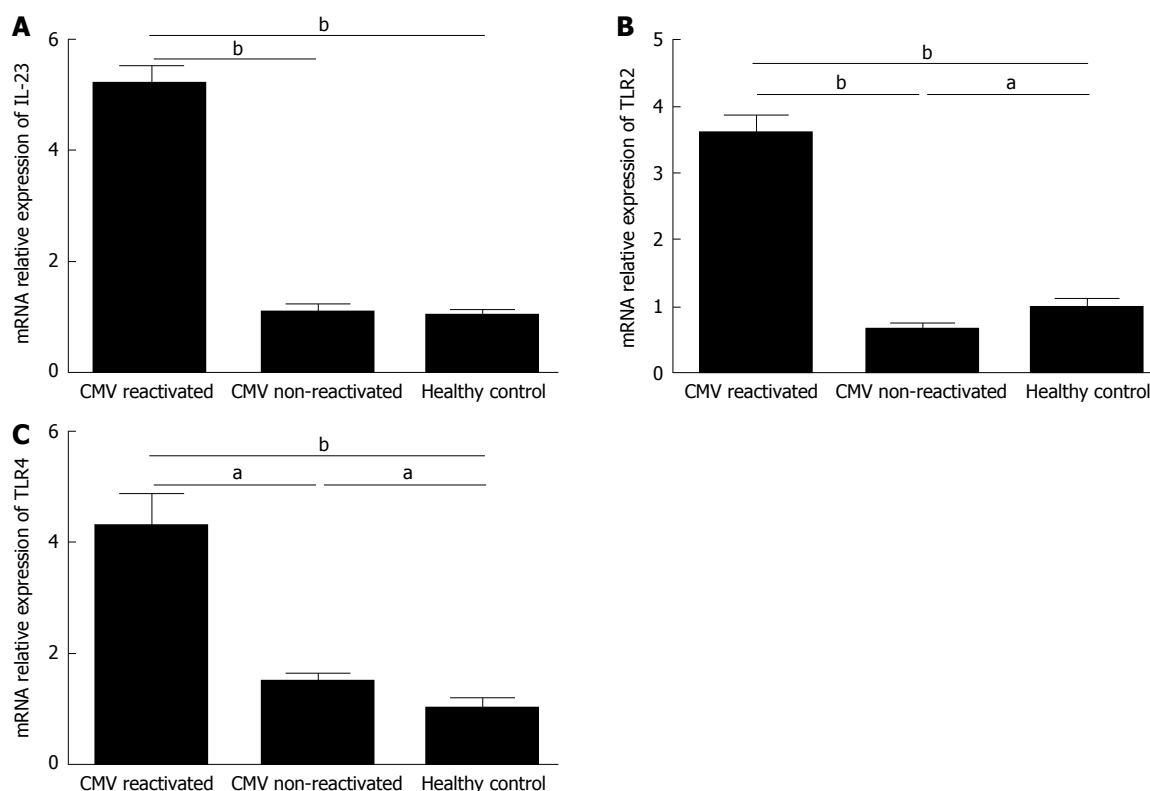
COMMENTS

Background

Cytomegalovirus (CMV) is the most determinative viral infectious pathogen cause of morbidity and mortality post-liver engraft and associate with diminished graft survival. CMV infected monocyte derived dendritic cells (CMV-MoDCs) have an altered phenotype and functional defects. DCs are determinative initiators of cellular immunity against CMV infection. CMV can interfere with maturation and antigen-presenting function of DCs and also disturb both innate and adaptive immunity. DCs undergo lytic viral cycles, can induce late gene expression of CMV, release of infectious virus, and stimulating of T-cell responses resulted to allograft rejection. However, association between CMV pathogenesis with DC maturation and function in CMV reactivated liver transplant patients was not yet evaluated.

Research frontiers

CMV can interfere with maturation and antigen-presenting function of DCs and also disturb both innate and adaptive immunity. This interference can promote viral spread by paralyzing the adaptive immune system. CMV with DC infection induce the production of inflammatory cytokines and activation of the interferon pathway in transplanted patients.

Innovations and breakthroughs

This is the first study evaluating the interference between CMV reactivation with maturation and antigen-presenting function of DCs in Iranian liver transplanted patients.

Applications

Interference of CMV and DCs can promote viral spread by paralyzing the adaptive immune system and induce the production of inflammatory cytokines and activation of the interferon pathway in transplanted patients. Results of this study highlight the fact that, CMV and DCs contractions promote different pathways including: Interference with the maturation and expression of DC markers and cytokines in MoDCs of CMV reactivated liver transplanted patients. These pathways can implicate in the development of acute or chronic allograft liver rejection.

Terminology

CMV infected MoDCs have an altered phenotype and functional defects. DCs are determinative initiators of cellular immunity against CMV infection. DCs also act with superiority over other antigen-presenting cells in stimulating T-lymphocyte responses and maintaining protective anti-CMV immunity.

Peer-review

The manuscript entitled, "Role of cytomegalovirus on the maturation and function of monocyte derived dendritic cells of liver transplant patients", by Karimi *et al*, demonstrated the functional defects of dendritic cells in CMV reactivated liver transplant recipients when compared to those without CMV reactivation or healthy norms demonstrating the differences in cytokine concentrations and expressions. This is very detailed investigation of cytokines of monocyte derived dendritic cells.

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