Effect of Influenza A Infection on Umbilical Cord Blood Natural Killer Function Regulation With Interleukin-15

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Background. Influenza A is a major pathogen of humans and has the potential to cause worldwide pandemics. Natural killer (NK) cells are important effector cells in the innate immune response against viruses, including influenza A. Infants are more susceptible to severe influenza A viral infection, possibly attributed in part to their defective NK function.

Methods. We compared the NK responses to influenza using umbilical cord blood (UCB) and adult peripheral blood (APB) mononuclear cells and purified NK cells.

Results. Influenza A induced dose-dependent apoptosis of NK cells with down-regulation of NKp46 expression, which was more pronounced in UCB. Both UCB and APB NK cells responded to influenza infection by up-regulating CD69 and CD107a expression, a process further enhanced by interleukin (IL) 15. Influenza exposure also down-regulated perforin expression and K562 cytototoxicity in UCB NK cells, which was partially restored by IL-15. The production of interferon (IFN) γ and tumor necrosis factor (TNF) α by NK cells in responding to influenza was further enhanced by IL-15.

Conclusions. Our findings show differential NK responses between newborns and adults. IL-15 may be beneficial in combating influenza by enhancing cytotoxic function and IFN-γ production.

Influenza is an acute respiratory viral infection that continues to pose endemic and pandemic threats to human health, with significant morbidity and mortality [1, 2]. Studies in humans and mice have implicated adaptive immune responses, including antibody and T-cell responses, in protective immunity against influenza infection [3, 4]. Studies in the mouse model have suggested that natural killer (NK) cells are also involved in the control of influenza [5, 6]; however, the role of NK cells in protective immunity in humans against influenza is poorly understood.

NK cells are key effector cells in innate immunity. They play a critical role in the first line of host defense against acute viral infections by directly destroying infected cells without the need for prior antigen stimulation [7, 8]. NK cells also exert an immunoregulatory function by modulating subsequent adaptive immune responses through crosstalk with dendritic cells [9, 10]. During the first few days of infection, influenza virus adapts to evade host innate immune responses, including NK cell immunity [11]. Recent studies showed that influenza virus directly infects NK cells, resulting in apoptosis [12] and decreased cytotoxic function [13]. Because of immunological naiveté and immaturity, children aged <24 months have a high risk of influenza-related hospitalization and morbidity [14, 15]. Young infants <6 months of age have the highest influenza-associated mortality rates [16]. Neonates are unusually susceptible to severe viral infections, such as influenza and human immunodeficiency virus (HIV), because of defective NK cell function [17, 18]. We and others have...
demonstrated that umbilical cord blood (UCB) NK cells exhibit lower expressions of adhesion molecules (CD2, CD11a, CD18, CD16, and CD54), compared with their adult counterparts, which may contribute to their deficient cytotoxic and regulatory functions [19, 20].

NK cells are activated during the initial stages of viral infections by cytokines and chemokines, including interferon (IFN) α, IFN-β, interleukin (IL) 12, IL-15, and IL-18, which are produced by infected cells or by activated dendritic cells and macrophages [21, 22]. IL-15 is an IL-2–like γ-chain signaling cytokine that stimulates viral-induced proliferation of NK cells [23] and enhances NK and memory CD8 T function in HIV-infected persons [24, 25]. It has unique antiapoptotic property distinct from IL-2 [26] and is critically involved in NK–dendritic cell interaction during acute viral infection [27, 28]. This makes IL-15 seem to be an attractive candidate for immunotherapy against influenza.

We postulated that deficient NK response in neonates might account for their greater susceptibility to influenza. In the present study, we compared the effects of influenza A virus infection on activation and apoptosis of NK cells from UCB and adult peripheral blood (APB). We also sought to determine whether IL-15 decreased NK apoptosis, augmented NK cytotoxic response, or enhanced productions of IFN-γ or tumor necrosis factor (TNF) α during in vitro influenza A virus infection.

**MATERIALS AND METHODS**

**NK Cell Preparation**

Mononuclear cells (MNCs) were isolated from heparinized APB or UCB samples with use of Ficoll-Hypaque density gradient centrifugation. Informed consent was obtained from each participant, and the study was approved by the Medical Ethics and Human Clinical Trial Committee of the Chang Gung Memorial Hospital. UCB was collected in sterile tubes and was processed within 24 hours after birth. MNCs were then resuspended in Roswell Park Memorial Institute (RPMI) medium with 10% fetal calf serum (FCS) at a concentration of 1 × 10^6 cells/mL.

NK cells were enriched from MNCs by depleting CD3^+ cells (T lymphocytes), CD19^+ cells (B lymphocytes), and CD14^+ cells (macrophage/monocytes) with use of the MACS Cell Isolation Kits for NK cells (Miltenyi Biotec), including bead-coupled monoclonal antibodies (mAbs) specific for CD3, CD4, CD19, and CD33. Through a MACS magnetic separator, NK cells were phenotypically enriched to consist of >90% CD16^+ /CD56^+ cells, as determined by flow cytometry. For subsequent experiments, MNCs or bead-purified NK cells were resuspended at a concentration of 1 × 10^6 cells/mL in RPMI with 10% FCS, with or without 10 ng/mL human recombinant IL-15 (Pepro Tech EC).

**Preparation of Influenza A Virus**

Influenza A/William Smith Neurotropic (WSN) virus/33 strain viruses were propagated in Madin–Darby canine kidney cells in Dulbecco’s modified Eagle medium (DMEM) with 2% FCS. Alternatively, allantoic fluid was harvested 48 hours after infection, and the concentration of influenza virus was measured. Virus-containing allantoic fluid was pooled and centrifuged to pellet viral particles. The virus pellets were resuspended in phosphate-buffered saline and further purified by centrifugation in a continuous of 15%–60% sucrose gradient. The purified virus was reconstituted in phosphate-buffered saline, stabilized with sucrose-phosphate-glutamate (BioWhittaker), dispensed into single-use aliquots, and stored at −70°C. The virus titer was determined with Madin-Darby canine kidney cells by standard procedures [29].

**Exposure of NK Cells to Influenza A Virus**

MNCs or purified NK cells were exposed to influenza A virus at a multiplicity of infection (MOI) of 0.1 or 1.0, for 1 hour at 37°C in serum-free RPMI 1640 supplemented with 2 mmol/L glutamine, 100 U of penicillin G, and 100 μg/mL streptomycin. Corresponding cells were sham-exposed (ie, cells were exposed to a volume of DMEM-cultured fluid equal to that used for virus infection). After 1 hour of viral or sham exposure, the MNC or purified NK cells were washed and incubated, with or without IL-15.

**NK Surface Marker Analysis and Intracellular Staining**

Cultured MNCs or NK cells were stained with fluorescein iso-thiocyanate (FITC)– or phycoerythrin (PE)–conjugated mouse anti-human mAbs, including anti-CD3/CD16CD56 (FITC/PE), CD69, and NKp46 (Becton-Dickinson) for cytometric analysis. Intracellular expression of NKp46, granzyme A, or perforin in NK cells was investigated after permeabilization of the cell membrane with a Cytofix/Cytoperm kit (BD Pharmingen) using FITC-conjugated corresponding antibodies. The fluorescent staining was analyzed on a FACScalibur (BD Biosciences) flow cytometer. Electronic gates were set to enable fluorescence analysis of the fluorescence of the viable cell population according to FSC/SSC (Forward Scatter/Side Scatter) histograms after anti-CD3 stimulation. The percentages of cells stained with each mAb were determined by comparing each histogram with one from control cells stained with FITC- or PE-labeled isotype control mAbs.

**Quantitative Real-Time Polymerase Chain Reaction for NKp46 RNA Expression**

Total RNA from mock-treated and influenza A–infected UCB and APB NK cells was extracted using Purelink Mocto-to-Midi Total Purification Kit (Invitrogen). The conversion of complementary DNA and real-time polymerase chain reaction (PCR) was then performed using the Power SYBR Green RNA-to-Ct 1-step kit (Applied Biosystems) and detected by the Bio-Rad iQ5 system. The primers used for NKp46 were as follows: forward, CTGAGCGATGTCTTCCACAC; reverse,
CCGCCAGGCTCAAC. The glyceraldehyde 3-phosphate dehydrogenase (GAPDH) gene was applied as a positive control. For qualification, the relative threshold cycle method (ΔΔCT) method) was applied. Data were analyzed using Bio-Rad iQ5 software.

Detection of Apoptosis
Influenza-exposed MNCs and NK cells were cultured without or with 10 ng/mL IL-15. The apoptosis levels were measured using the annexin V and propidium iodide (PI) detection method, as described elsewhere [30]. The fluorescent staining was analyzed using a FACSCalibur flow cytometer (BD Biosciences).

Figure 1. Effect of influenza (fluA) virus infection on percentage of CD69 expression of CD3⁻CD56⁺ natural killer (NK) cells in mononuclear cells (MNCs) (A) and magnetic bead-purified NK cells from adult peripheral blood (APB) and umbilical cord blood (UCB) (B). Purified NK cells were also incubated in the absence (media) or presence of 10 ng/mL interleukin (IL) 15 for 24 hours before infection. Data were obtained from 11 adult and 14 cord samples. For multiple comparison of 2 doses of fluA, differences were considered statistically significant only at \( P < .03 \). \* \( P < .05 \), \*\* \( P < .01 \) for 2 groups being compared. Values are expressed as mean percentage ± standard error of the mean (SEM). Abbreviation: MOI, multiplicity of infection.

CD107a Degranulation Assay
The degranulation assay was performed as described elsewhere [31], with minor modifications. MNCs and purified NK cells were stained with anti-CD107a FITC and CD56 PE (BD Biosciences) and incubated for 25 minutes at 37°C in the dark. In some experiments, purified NK cells were cocultured for 4 hours with K562 cells at ratios of 2.5:1 and 1:1 and then analyzed on a FACSCalibur flow cytometer.

NK Cells Cytotoxic Function
Flow cytometric NK cytotoxicity assays were performed as described elsewhere [32]. In brief, purified unstimulated or IL-15–treated NK cells (effectors) and the erythromyelocytic
leukemia cell line K562 cells (targets) were added in a tube. Control tubes including only target or effector cells were assayed to determine the spontaneous cell death. After the incubation, CD45-FITC (BD Bioscience) was added and incubated for 20 minutes on ice. Then PI was added to each tube 10–15 minutes before acquisition. The percentages of dead K562 cells were calculated on the basis of PI uptake of K562 cells after subtraction of background cell death.

**Measurement of IFN-γ and TNF-α Protein Expression**

Secreted IFN-γ or TNF-α from purified UCB and APB NK cells was determined in culture supernatants using a human IFN-γ or human TNF-α enzyme-linked immunosorbent assay kit (R&D Systems), as recommended by the manufacturers. For the intracellular IFN-γ staining, purified NK cells with different treatments were stimulated for 4 hours with phorbol 12-myristate 13-acetate (PMA, 50 ng/ml) and ionomycin (1 μg/mL) in the presence of GolgiStop protein transport inhibitor.
RESULTS

Up-Regulation of CD69 Expression of NK Cells by Influenza Virus
We first examined the expression of the early activation marker CD69 in NK cells that were cocultured with influenza A viruses. Figure 1A shows CD69 expression on UCB and APB NK cells (CD3+/CD56+) in bulk MNC cultures (Figure 1A). Influenza A exposure resulted in a dose-dependent increase of CD69 expression on both UCB and APB NK cells, with levels of UCB NK cells exceeding those on corresponding APB NK cells (for 0.1 MOI, 36.8% ± 3.4% vs 18.4% ± 2.1% [P = .002]; for 1 MOI, 44.4% ± 1.9% vs 21.9% ± 3.1% [P = .003]).

We next compared the effect of IL-15 on CD69 expression in bead-purified NK cells exposed to influenza A at an MOI of 1 (Figure 1B). IL-15 enhanced CD69 expression of mock-treated UCB NK cells and corresponding APB NK cells to similar extents (58.1% ± 5.4% vs 79.5% ± 7.6% [P = .003]). Influenza A exposure also resulted in enhanced CD69 expressions in bead-purified UCB and APB NK cells (24.0% ± 2.8% vs 31.0% ± 2.9% [P = .189]). IL-15 further enhanced similar levels of CD69 expression on APB (P = .003) and UCB (P = .001) NK cells after influenza virus infection.

Influenza Exposure and Down-Regulation of NKP46 Expression in NK Cells
Because NKP46 might be the receptor for influenza hemagglutinin, we observed that influenza A did bind to NK cells. Although UCB NK cells expressed higher levels of NKP46 in purified NK cells (Figure 2A and Table 1), there was a greater decrease of NKP46 expression in UCB NK cells after influenza A exposure than in APB NK cells (Figure 2B).
APB NK cells (Figure 2B). IL-15 did not influence the NKp46 expression of UCB and APB NK cells, regardless of infection status. To rule out the possibility that the down-regulation of NKp46 was attributable to the blocking of antibody by influenza A, intracellular staining (Figure 2C) and real-time PCR quantifying messenger RNA (data not shown) were also performed. The results confirmed that influenza A indeed reduced intracellular NKp46 protein and RNA expression in NK cells.

**Influenza Exposure and Induced Apoptosis of NK Cells**

MNC exposure to influenza A resulted in similar levels of dose-dependent apoptosis of UCB and APB NK cells (for 0.1 MOI, 34.9% ± 4.3% vs 44.7% ± 5.5% [P = .303]; for 1 MOI, 47.2% ± 6.2% vs 45.8% ± 4.5% [P > .99]) (Figure 3A). When purified APB or UCB NK cells were analyzed, the percentages of annexin V^+/PI^- (early apoptotic cells) NK cells were higher than in corresponding mock-treated controls (P < .01) (Figure 3B). UCB NK cells were more susceptible...
Figure 4. A, Representative profile showing the effect of influenza A (fluA) virus infection on CD107a degranulation of purified adult peripheral blood (APB) natural killer (NK) cells after contact with K562 cells and the effect of interleukin (IL) 15. The combination of fluA infection and IL-15 produced the greatest increase in CD107a expression. B, C, Percentage of CD107a expression of CD3⁻CD56⁺ NK cells in mononuclear cells (MNCs) (B) or magnetic bead–purified NK cells (C) from adult peripheral blood (APB) and umbilical cord blood (UCB). Purified NK cells were also incubated in the absence (media) or presence of 10 ng/mL IL-15 for 24 hours before infection. Data were obtained from 11 adult and 14 cord samples. For multiple comparison of 2 doses of fluA, differences were considered statistically significant only at $P < .03$. *$P < .05$, **$P < .01$ for 2 groups being compared. Values are expressed as mean percentage ± standard error of the mean (SEM). Abbreviation: MOI, multiplicity of infection.
Figure 5. A, B, Effect of influenza A (fluA) virus infection on K562 cytotoxicity of mononuclear cells (MNCs) (A) and primary purified natural killer (NK) cells (B) from adult peripheral blood (APB) and umbilical cord blood (UCB). C, D, Mean fluorescence intensity (MFI) of granzyme (C) and perforin (D)
Effect of Influenza Exposure and IL-15 on CD107a Degranulation of NK Cells

Figure 4 shows a representative profile of the effect of influenza A on CD107a degranulation of purified APB NK cells after contact with K562 cells and the effect of IL-15. Influenza-exposed MNCs showed a dose-dependent increase of CD107a expression in both APB (for MOI of 0.1, 8.1% ± 1.9% [P = .043]; for MOI of 1, 11.0% ± 3.4% [P = .043]) (Figure 4B) and UCB NK cells (for MOI of 0.1, 8.2% ± 1.2% [P = .028]; for MOI of 1, 17.2% ± 4.2% [P = .043]). Using purified NK cells (Figure 4C), we observed an up-regulation of CD107a expression by influenza A in APB (P = .003) and more pronounced in UCB (P = .004) (Table 1). IL-15 further enhanced CD107a expression and had significant effect on mock-treated UCB NK (P = .035) and influenza A virus–infected APB NK cells (P = .013).

Influenza Viral Infection and Down-Regulation of Cytotoxic Function of NK Cells

When NK function was assessed using influenza A–exposed MNCs, the cytotoxicity toward K562 cells was lower in UCB than in APB (Figure 5A). Influenza A exposure resulted in a slightly dose-dependent increase in K562 cytolyis of UCB MNCs (for MOI of 0.1, 6.2% ± 1.6% [P = .043]; for MOI of 1, 13.7% ± 2.4% [P = .043]), but not in APB. However, when bead-purified NK cells were used to assess the direct effect of influenza A on cytotoxicity, we found that an MOI of 1 significantly down-regularized the K562 cytotoxicity of bead-purified APB NK cell (17.9% ± 4.2% vs 26.7% ± 6.1%; P = .034) and UCB NK cells (8.4% ± 2.2% vs 14.7% ± 4.6%; P = .021) (Figure 5B). IL-15 significantly enhanced the K562 cytotoxicity of influenza virus–infected APB (45.3% ± 5.3%; P = .003) and UCB NK cells (45.3% ± 5.2%; P = .001). The level of K562 cytotoxicity of influenza A–treated IL-15 activated APB and UCB NK cells was still lower than that in corresponding mock-treated APB (70.61% ± 3.73%; P = .003) and UCB (67.1% ± 6.5%; P = .001), respectively.

We also examined the effect of influenza A on cytolytic machinery by measuring the granzyme A and perforin expressions of purified APB and UCB NK cells. We observed higher granzyme A expression in UCB NK cells (MFI, 20.7 ± 3.5) than in APB NK cells (MFI, 10.1 ± 1.2; P = .01) (Figure 5C). However, influenza A exposure did not affect the level of granzyme A expression of APB (P = .86) and UCB NK cells (P = .78), whereas IL-15 uniformly enhanced granzyme A expression. In contrast, influenza A significantly inhibited perforin expression in UCB NK cells (MFI, 88.2 ± 14.5 vs 118.8 ± 18.1; P = .041) (Figure 5D). Similar to the observations with granzyme A, IL-15 enhanced perforin expression on influenza A–infected APB (P = .033) and UCB NK cells (P = .001).

Effect of Influenza on IFN-γ and TNF-α Production of NK Cells

We next examined the effects of influenza A exposure on IFN-γ and TNF-α production of purified NK cells. Although the intracellular staining indicated that the percentages of IFN-γ+ APB or UCB NK cells were not changed after influenza A virus infection (Figure 6A), influenza A induced higher levels IFN-γ in the culture supernatants of APB (74.5 ± 29.5 pg/mL) and UCB (103.8 ± 33.5 pg/mL) NK cells, compared with mock-treated controls (Figure 6B). IL-15 further enhanced the IFN-γ production of influenza A–treated NK cells in APB (335.5 ± 96.6 pg/mL; P = .018) and UCB (586.4 ± 155.4 pg/mL; P = .005) alike. Similar to the observations with IFN-γ production, IL-15 significantly enhanced TNF-α production of influenza A–infected APB and UCB NK cells (Figure 6C).

DISCUSSION

Previous studies have had controversial results regarding effects of the influenza virus on NK cells. Some studies demonstrated an NK-activating effect [23, 33], whereas several recent studies showed a detrimental effect of influenza A on NK cytotoxic function [12, 34, 35]. The present study examined the effects of influenza A on various aspects of NK function by gating NK cells in MNCs or bead-purified NK cells. Comparison of the responses of NK cells from adult and cord blood led us to speculate that neonatal NK cells might be deficient in combating influenza A and that NK-enhancing cytokines, such as IL-15, may provide some resistance in the context of influenza virus infection.

After influenza exposure, we observed a dose-dependent increase of CD69 expression on NK cells in MNCs, in agreement with Du et al [33]. We further showed that the UCB NK CD69 response exceeded that in corresponding adult cells. When purified NK cells were compared, however, the UCB response was comparable to that of APB. This discrepancy...
Figure 6.  

A. Effect of influenza A (fluA) virus infection on the ratio of interferon (IFN) γ⁺ cells, based on the intracellular cytokine staining and compared with the percentage of each media culture.  

B, C. With enzyme-linked immunosorbent assay (ELISA), the levels of IFN-γ (B) and tumor necrosis factor (TNF) α (C) in magnetic bead–purified natural killer (NK) cells from adult peripheral blood (APB) and umbilical cord blood (UCB) were also determined. Purified NK cells were incubated in the absence (media) or presence of 10 ng/mL interleukin (IL) 15 for 24 hours before infection. Data were obtained from 6 adult and 6 cord samples for IFN-γ intracellular cytokine staining and 9 adult and 10 cord samples for ELISA. *P < .05, **P < .01. Values are expressed as mean concentration (picograms per milliliter) ± standard error of the mean (SEM).
may be attributable to the effect of influenza on dendritic cells or monocytes in MNCs, which subsequently activate NK cells by direct contact or secreting cytokines. In addition, we observed that IL-15 augmented the CD69 expressions of influenza A–infected UCB and APB NK cells. Our findings suggest that neonatal NK cells were not deficient in up-regulating CD69 in response to influenza A.

NKp46 is the major triggering receptor involved in the natural cytotoxicity of fresh or cultured human NK cells and binds influenza virus hemagglutinin [36]. We confirmed the results of previous studies [33, 37] showing that NKp46 expression was down-regulated in a dose-dependent manner by influenza A. This effect was not attributable to the blocking of antibody by influenza A, confirmed with intracellular staining and real-time PCR. However, we are the first to show a greater decrease in NKp46 expression in UCB than in APB, suggesting that UCB NK cells may be more susceptible to the detrimental effects of influenza A. IL-15 had little effect on NKp46 expression in mock-treated or influenza virus–infected NK cells.

We confirmed the findings of Mao et al [13] that influenza A infected NK cells and induced apoptosis. These findings partially explain the clinically observed decrease in circulating NK cells during severe influenza virus infection [38]. We further showed that influenza A–induced apoptosis in UCB NK cells was more pronounced than in APB. Thus, a deficient NK defense against influenza may contribute to the increased hospitalization of infants <6 months of age [14, 15], despite the possible protection provided by maternal antibodies. Although considered to be an important survival factor for NK cells, IL-15 did not decrease influenza-induced NK apoptosis.

Lysosomal-associated membrane protein–1 (or CD107a) has been described as a marker in NK cell degranulation after stimulation and may represent a good marker for NK functional activity [31]. We observed an enhanced CD107a expression in both NK cells in influenza A–exposed MNCs and purified NK cells, in agreement with Du et al [33]. The CD107a expression was further enhanced after contact with K562 cells, a process that was facilitated by IL-15, confirming a previous report [39]. We showed that UCB NK cells responded to IL-15 more readily than do their adult counterparts with regard to CD107a degranulation after contact with K562 cells and influenza A.

Others and we have shown that UCB NK cells are deficient in cytotoxic function but readily respond to IL-15 stimulation [32, 40]. UCB MNC exposure to influenza A resulted in enhanced K562 cytotoxicity, whereas in purified NK cells, we observed a detrimental effect of influenza A on NK cytotoxicity. This discrepancy partially explains the reported variation in the effects of viral infection on NK function and highlights the importance of obtaining purified NK cells. To our knowledge, we are the first to describe deficient perforin expression in UCB NK but not APB NK cells after influenza virus infection.

Dampening NK cytotoxic function is a novel strategy used by the influenza virus to evade NK cell innate immune defense and thereby facilitate viral transmission [11, 34]. IL-15 seems to be beneficial in this regard, because it effectively enhanced K562 cytotoxicity in influenza A virus–infected UCB and APB NK cells.

Weiss et al showed that IFN-γ treatment at early stages of influenza virus infection protect mice from death in a NK cell–dependent manner [41]. In agreement with findings of previous studies of intracellular cytokine staining and influenza H3N2 virus infection [42, 43], we did not detect enhanced percentages of IFN-γ+ cells after WSN influenza A H1N1 virus infection. However, some levels of IFN-γ in culture supernatants were detected by influenza A, which was also observed by another study with influenza PR8 H1N1 [44]. The discrepant results of the 2 techniques could be attributable to different sensitivities between assays and different virus strains used. Small et al [35] reported that TNF-α production of NK cells played an important role in NK cell–mediated antibacterial host defense. We found that IL-15 could further enhance IFN-γ and TNF-α production of influenza virus–infected NK cells, with UCB response exceeding that of APB. Administering IL-15 may be particularly of therapeutic benefit in newborns with overwhelming influenza virus infection. In conclusion, our finding showed that the differential NK response to influenza virus infection between newborn and adults. IL-15 may be beneficial in combating influenza by enhancing cytotoxic function and augmenting IFN-γ and TNF-α production.

Notes

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References