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HIV/HCV Co-Infection Is Associated With Significant Alterations of the NK Cell Pool

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Background

Hepatitis C virus (HCV) co-infection in HIV(+) patients is associated with faster progression of liver disease as compared to HCV mono-infection. Natural killer (NK) cells critically modulate the natural course of hepatitis C. Of note, chronic infection with HCV as well as HIV mono-infection have been shown to be associated with significant alterations of the NK cell pool. However, little data is available concerning phenotype and function of NK cells in HIV/HCV co-infection.

Methods

A total of 22 HIV/HCV co-infected patients were enrolled into this study. As control, 70 patients with chronic hepatitis C, 39 HIV mono-infected patients and 55 healthy individuals were analyzed. NK cell phenotype as well as IFN-γ production and degranulation (CD107a) of NK cells were studied by flow cytometry.

Table 1. Patient characteristics

	Healthy	HIV	HCV	HIV/HCV
Number	55	39	70	22
Male sex[a]	27 (49 %)	31 (79 %)	42 (62 %)	19 (86 %)
Age (years)[b]	30,8 (21 – 52)	44,2 (22 – 81)	45,6 (17 – 73)	44,3 (31 – 58)
Clinical data				
AST (U/L)[b]	n/a	30,8 (14 – 73)	63,2 (15 – 249)	73 (12 – 291)
ALT (U/L)[b]	n/a	32,7 (10 – 96)	97,5 (9 – 705)	78,3 (24 – 181)
gammaGT (U/L)[b]	n/a	76,4 (16 – 347)	104,8 (13 – 723)	124,9 (32 – 413)
Bilirubin (mg/dL)[b]	n/a	n/a	0,67 (0,23 – 1,7)	0,83 (0,26 – 2,76)
HIV status				
HIV RNA <40 copies/mL (under HAART)[a]	n/a	35 (90 %)	n/a	21 (95 %)
CD4 (cells/µL)[b]	n/a	555 (124 – 1199)	n/a	577 (182 – 1151)
CD8 (cells/µL)[b]	n/a	1115 (364 – 2163)	n/a	1000 (254 – 2249)
HCV status				
HCV RNA (IU/mL)[b]	n/a	n/a	3.026.784 (258 – 21.539.274)	3.479.717 (78.500 – 26.085.192)
HCV genotype				
Genotype 1[a]	n/a	n/a	53 (76 %)	16 (73 %)
Genotype 2[a]	n/a	n/a	0	0
Genotype 3[a]	n/a	n/a	14 (20 %)	0
Genotype 4[a]	n/a	n/a	1 (1,4 %)	4 (18 %)
more than one[a]	n/a	n/a	1 (1,4 %)	1 (4,5 %)
unknown[a]	n/a	n/a	1 (1,4 %)	1 (4,5 %)

a) number of cases (% of total)
b) mean (range)
n/a = not available / not analyzed

Results

Figure 1

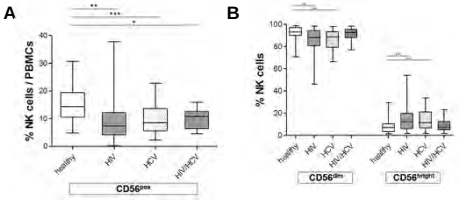


Fig. 1. Frequency of NK cells. (A) Frequency of total NK cells was significantly lower in co-infection than in healthy individuals (p=0.016) but did not differ from HIV and HCV mono-infection. **(B)** There was no significant difference between co-infected and healthy individuals with respect to the percentage of $CD56^{dim}$ and $CD56^{bright}$ subpopulations of total NK cells.

Figure 2

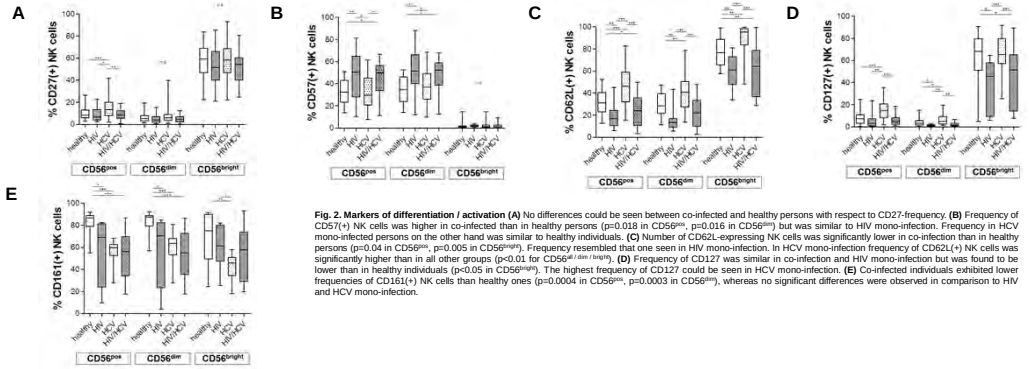


Fig. 2. Markers of differentiation / activation (A) No differences could be seen between co-infected and healthy persons with respect to CD27-frequency. **(B)** Frequency of CD57(+) NK cells was higher in co-infected than in healthy persons (p=0.018 in $CD56^{pos}$, p=0.016 in $CD56^{dim}$) but was similar to HIV mono-infection. Frequency in HCV mono-infected persons on the other hand was similar to healthy individuals. **(C)** Number of CD62L-expressing NK cells was significantly lower in co-infection than in healthy persons (p=0.04 in $CD56^{pos}$, p=0.005 in $CD56^{dim}$). Frequency resembled that one seen in HIV mono-infection. In HCV mono-infection frequency of CD62L(+) NK cells was significantly higher than in all other groups (p<0.01 for $CD56^{dim\,/\,bright}$). **(D)** Frequency of CD127 was similar in co-infection and HIV mono-infection but was found to be lower than in healthy individuals (p<0.05 in $CD56^{bright}$). The highest frequency of CD127 could be seen in HCV mono-infection. **(E)** Co-infected individuals exhibited lower frequencies of CD161(+) NK cells than healthy ones (p=0.0004 in $CD56^{pos}$, p=0.0003 in $CD56^{dim}$), whereas no significant differences were observed in comparison to HIV and HCV mono-infection.

Figure 3

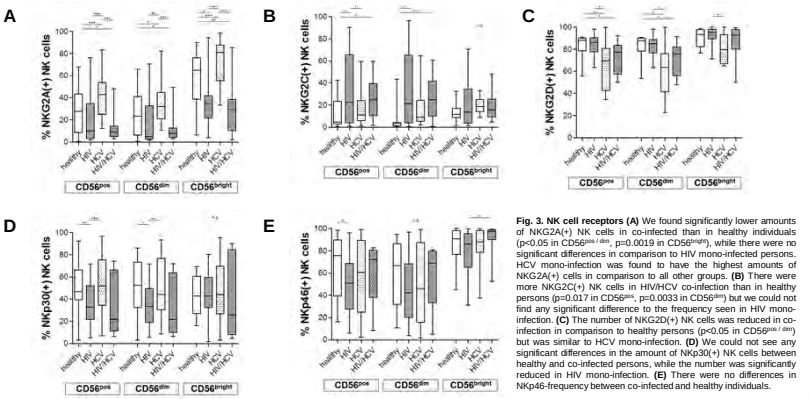


Fig. 3. NK cell receptors (A) We found significantly lower amounts of NKG2A(+) NK cells in co-infected than in healthy individuals (p<0.05 in $CD56^{pos\,/\,dim}$, p=0.0019 in $CD56^{bright}$), while there were no significant differences in comparison to HIV mono-infected persons. HCV mono-infection was found to have the highest amounts of NKG2A(+) cells in comparison to all other groups. **(B)** There were more NKG2C(+) NK cells in HIV/HCV co-infection than in healthy persons (p=0.017 in $CD56^{pos}$, p=0.0033 in $CD56^{dim}$) but we could not find any significant difference to the frequency seen in HIV mono-infection. **(C)** The number of NKG2D(+) NK cells was reduced in co-infection in comparison to healthy persons (p<0.05 in $CD56^{pos\,/\,dim}$) but was similar to HCV mono-infection. **(D)** We could not see any significant differences in the amount of NKp30(+) NK cells between healthy and co-infected persons, while the number was significantly reduced in HIV mono-infection. **(E)** There were no differences in NKp46-frequency between co-infected and healthy individuals.

Figure 4

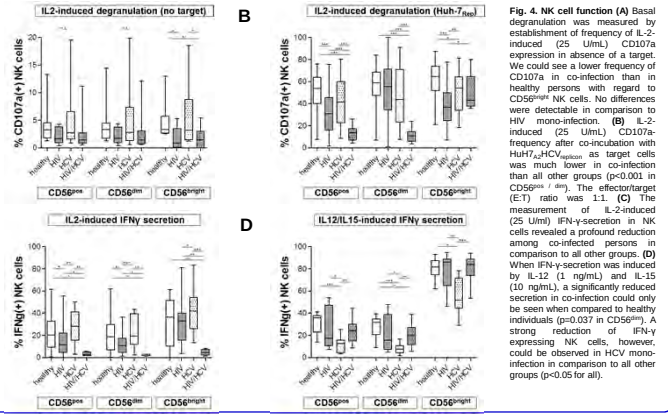


Fig. 4. NK cell function (A) Basal degranulation was measured by establishment of frequency of IL-2-induced (25 U/mL) CD107a expression in absence of a target. We could see a lower frequency of CD107a in co-infection than in healthy persons with regard to $CD56^{bright}$ NK cells. No differences were detectable in comparison to HIV mono-infection. **(B)** IL-2-induced (25 U/mL) CD107a-frequency after co-incubation with $HuH7.5cHCV_{replicon}$ as target cells was much lower in co-infection than all other groups (p<0.001 in $CD56^{pos\,/\,dim}$). The effector/target (E:T) ratio was 1:1. **(C)** The measurement of IL-2-induced (25 U/ml) IFN-γ-secretion in NK cells revealed a profound reduction among co-infected persons in comparison to all other groups. **(D)** When IFN-γ-secretion was induced by IL-12 (1 ng/mL) and IL-15 (10 ng/mL), a significantly reduced secretion in co-infection could only be seen when compared to healthy individuals (p=0.037 in $CD56^{dim}$). A strong reduction of IFN-γ expressing NK cells, however, could be observed in HCV mono-infection in comparison to all other groups (p<0.05 for all).

Conclusion

Our data indicate that HIV/HCV co-infection is associated with significant alterations of the NK cell phenotype and NK cell functions, which might be involved in the rapid progression of liver disease observed in co-infected patients. Of note, many NK cell alterations in co-infection resemble alterations as they can be seen in HIV mono-infected individuals, suggesting a pronounced impact of HIV on the phenotype of NK cells in the setting of co-infection.

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