Hot News

Interleukin 21, Essential to Control Chronic Viral Infections

HIV and hepatitis C (HCV) infections are two of the most relevant human chronic viral infections worldwide. They often coexist within the same host. Immune responses mediated by T lymphocytes are generally associated with control of viral replication. However, an impaired specific T-cell response is a hallmark of both HIV and HCV persistence. $CD4^+$ T-cell responses are required to sustain virus-specific $CD8^+$ T-cell functions during chronic viral infections. Therefore, the loss of expression of cytokines by virus-specific $CD4^+$ T-cells could be an important factor underlying the failure of $CD8^+$ T-cells to control these viral infections.

Abnormalities in the production of important antiviral and immunostimulatory cytokines, such as interleukin-2, interferon-gamma, and tumor necrosis factor-alpha, have been the major focus of attention until now. However, three recent reports (Elsaesser, et al. Science. 2009;324:1569-72; Yi, et al. Science. 2009;324:1572-76; and Fröhlich, et al. Science. 2009; 324:1576-80) have moved the attention to interleukin-21 (IL-21), a cytokine produced primarily by $CD4^+$ T-helper cells that could be the building block that allows $CD4^+$ T-cells to help $CD8^+$ T lymphocytes struggle against chronic viral infections. IL-21 promotes effective virus-specific T-cell responses through the IL-21 receptor (IL-21R)-dependent signaling in $CD8^+$ T-cells.

The new studies were performed on mice infected with the lymphocytic choriomeningitis virus (LCMV), which can cause acute or chronic infection depending on the viral strain and the size of the inoculum. Results from these studies highlight that IL-21 and IL-21R are not essential for efficient control of acute viral infection, but are crucial for sustained functionality of LCMV-specific $CD8^+$ T-cells and for the consequent control of chronic LCMV infection. A reduced production of IL-21 during the initial phase of chronic infection provoked the impairment of anti-LCMV $CD8^+$ T-cell responses and the failure to control viral infection. Also, IL-21R-deficient mice developed chronic viremia, even following exposure to low viral doses. Altogether, these findings support that a reduced production of IL-21 could be a distinctive characteristic of the loss of $CD4^+$ T-cell functionality commonly seen during the evolution of chronic HIV and HCV infections.

Two previous studies on HIV (White, et al. Blood. 2007;109:3873-80; and Strbo, et al. AIDS. 2008;22:1551-60) have demonstrated that IL-21 is a potent inducer of perforin in natural killer and $CD8^+$ T-cells, suggesting effects of IL-21 on both innate and adaptive immune responses. However, further research is necessary to prove the value of IL-21 in restoring the antiviral adaptive immune responses during HIV and HCV chronic infections.

Enhanced proliferation and increased cytotoxicity of $CD8^+$ T-cells mediated by IL-21 should be proved, evaluating IL-21 production by $CD4^+$ T-cells and its effect on $CD8^+$ T-cell function in patients experiencing HIV progression in comparison with subjects with HIV non-progression, as well as in chronic HCV infection compared to self-limited acute HCV infection.

Finally, the enhancing effector function of virus-specific $CD8^+$ T-cells during chronic infections is a quality of IL-21 that could be utilized for therapeutic uses. An immunotherapy based on IL-21 supply could be a new hope for HIV and HCV chronically infected patients, mainly for those in whom the current treatments had failed. Nevertheless, despite IL-21 having been safely used as antitumor immunotherapy in humans (Dodds, et al. Cancer Immunol. 2009;58:843-54), it has pleiotropic effects on both innate and adaptive immune responses, so it is important to assess its potential inhibitory effects on other immune cells before exploring application of this treatment strategy to chronic viral infections.

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Antiretroviral Drugs – Once Versus Twice a Day

Schedules of drug administration are normally based on the elimination half-life ($t_{1/2}$) of the drugs. Medications with long $t_{1/2}$ (> 24 hours), such as non-nucleoside reverse transcriptase inhibitors (NNRTI), can be dosed once a day. By contrast, most protease inhibitors (PI), nucleoside reverse transcriptase inhibitors (NRTI), and integrase strand transfer inhibitors (InSTI) have shorter $t_{1/2}$ and, therefore, are generally given twice a day. However, the particular pharmacodynamic and pharmacokinetic characteristics of NRTI and InSTI may also support once-a-day dosing.

The NRTI are prodrugs that must be converted intracellularly into their triphosphate forms to exert their antiviral effect. Since the intracellular $t_{1/2}$ of almost all triphosphate anabolites is longer than the plasma $t_{1/2}$ of their corresponding prodrugs, once-a-day dosing may be advisable in most cases. However,

thymidine analogs, such as zidovudine (AZT) and stavudine (d4T), display 3-4 hour intracellular $t_{1/2}$, which would not support once-daily dosing, and accordingly they are recommended to be prescribed twice a day. In the case of abacavir (ABC), which also displays a relatively short intracellular $t_{1/2}$, *in vitro* studies have demonstrated that it can be given once a day (Drusano, et al. Antimicrob Agents Chemother. 2002;46:464-70). In this study, the once-daily and twice-daily doses provided the same suppressive effect compared to continuous infusion, as evaluated using the daily area under the concentration-time curve ($AUC_{0\text{-}24h}$). Several hypotheses have been proposed to explain this observation, including the following: (i) a Michaelis-Menten step in the anabolism of ABC to carbovir triphosphate; (ii) an underestimation of the triphosphate $t_{1/2}$; and (iii) the existence of a post-antiviral effect resembling what is known as post-antibiotic effect for some antimicrobial agents (e.g. aminoglucosides).

In vivo and *in vitro* pharmacokinetic-pharmacodynamic studies of raltegravir have suggested that $AUC_{0\text{-}24h}$ and not trough concentrations (C_{trough}) is the most relevant pharmacokinetic parameter describing the antiviral activity of the drug (Wenning, et al. 9th International Workshop on Clinical Pharmacology of HIV Therapy. 7-9 April 2008, New Orleans, abstract O21; McSharry, et al. 10th International Workshop on Clinical Pharmacology of HIV Therapy. 15-17 April 2009, Amsterdam, abstract O_09). The *in vitro* study compared equivalent exposures of raltegravir (same $AUC_{0\text{-}24h}$), either in a continuous infusion mode or as fractionated oral doses in three different schedules (every 8, 12, and 24 hours). Overall, there were no significant differences in the suppressive effect. The explanation for this pharmacokinetic dependence of $AUC_{0\text{-}24h}$ was based on the interesting finding that the residence time of InSTI on the integrase/DNA complex was comparable or exceeding the $t_{1/2}$ of the pre-integration complex within the cell, resulting in a functionally irreversible inhibition of integration (Grobler, et al. 10th International Workshop on Clinical Pharmacology of HIV Therapy. 15-17 April 2009, Amsterdam; abstract O_10).

Altogether, these data are consistent with the recent finding of the crucial role played by the dose-response curve slope on the antiviral activity of antiretroviral drugs (Shen, et al. Nat Med. 2008; 14:762-6). This new parameter, which represents a measure of cooperation in the binding of multiple ligands to linked binding sites, is class-specific for antiretroviral drugs as it depends of the mechanism of action. The slopes for all NRTI and InSTI are ~ 1, a typical feature of non-cooperative reactions, whereas the slopes for NNRTI and PI are > 1, reflecting cooperative reactions. Thus, continuous drug exposure is not required for NRTI and InSTI in order to provide viral inhibition since the effect on the complex "enzyme/viral genome" is irreversible. Accordingly, the efficacy of NRTI and InSTI is $AUC_{0\text{-}24h}$ and not C_{trough} dependent, supporting that once-a-day prescription should be as effective as twice-a-day administration. In contrast, the existence of an intermolecular cooperation for NNRTI and PI may require continuous drug exposure over time to keep the enzyme inhibited. Accordingly, C_{trough} is the best parameter reflecting the virologic inhibitory effect of NNRTI and PI. Only if their $t_{1/2}$ is long enough, may they be given once a day.

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