Dilated cardiomyopathy, one of the leading causes of heart failure in the United States, is a multifactorial disease that includes both hereditary and acquired forms. In patients, it has been shown that dilated cardiomyopathy can be a sequela of viral myocarditis. Although many different infectious agents have been attributed as the cause of viral myocarditis, enteroviruses, in particular Coxsackie B viruses, are consistently among the most common. The concept that enteroviruses contribute to the pathogenesis of a subset of human dilated cardiomyopathy has been strengthened by the detection of enteroviral genome in the hearts of patients with dilated cardiomyopathy. Since the first description by Bowles et al in 1986, many articles have addressed this issue (reviewed in Baboonian et al). Although the results of individual studies vary, the data overall indicate that enteroviral genome is present in the heart of 15% to 25% of patients with dilated cardiomyopathy.

Analogous to many other viral illnesses, both direct viral injury and the immune response of the host play an important role in the pathogenesis of viral heart disease. Furthermore, results from experiments in murine models of viral myocarditis indicate that although the immune response has an important protective role, it may also have deleterious effects on the host. The balance between these protective and deleterious effects may ultimately determine the course of disease after enteroviral infection.

The direct viral effects have been demonstrated in culture and in vivo. In cultured cardiomyocytes, infection with Coxsackievirus B3 (CVB3) induces a direct cytopathic effect and cell death. In mice, transgenic expression of CVB3 genomes in the heart is sufficient to induce dilated cardiomyopathy. Both effects occurred independently of an immune response and demonstrate that coxsackieviral proteins can principally cause myocyte damage and heart disease. In this regard, a molecular pathogenic mechanism has been proposed that involves dystrophin, a cytoskeletal protein that causes dilated cardiomyopathy when genetically defective. Coxsackieviral protease 2A proteolytically cleaves and functionally impairs dystrophin during CVB3 infection.

In an immunocompetent host, the immune response elicited by viral proteins recognized as foreign limits viral replication and, in many cases, finally clears the virus from the host. These antiviral immune properties are highlighted by experiments using immunocompromised mice infected with CVB3. Mice with severe combined immunodeficiency and mice treated with the immunosuppressive agent FK506 demonstrate enhanced myocardial damage in the absence of an effective immune response. In addition, knockout of the inducible nitric oxide synthase gene (NOS2) that is expressed at high levels in immune cells causes increased viral titers and myocarditis in CVB3-infected mice. These studies clearly show the protective aspects of mononuclear cells in murine myocarditis and the potential for direct viral mediated cardiac injury.

However, the immune response may also contribute to tissue damage by inappropriately attacking cardiac myocytes. In 1974, Woodruff and Woodruff first demonstrated a role for T lymphocytes in the pathogenesis of murine myocarditis when they showed that depletion of T lymphocytes using antithymocyte serum or thymectomy and irradiation led to a decrease in mortality and a decrease in the inflammatory infiltrate after CVB3 infection. Subsequently, considerable research has been performed to determine the role of T-cell subtypes in the immunopathogenesis of viral myocarditis. This includes work by Huber et al that has demonstrated an important role for Th1 cell responses and the requirement for γδ T-cell activation.

It has been proposed that molecular mimicry—the presence of related epitopes—exists between CVB3 and a variety of myocardial proteins. This can lead to antibodies or cytotoxic T lymphocytes originally directed against the virus to cross-react with host antigens. These harmful immune properties were demonstrated by transfer of mononuclear cells from mice infected with CVB3 or patients with myocarditis into genetically identical or immunodeficient mice, respectively. In addition, increased levels of cytokines, such as tumor necrosis factor (TNF)-α, may also contribute to the decrease in myocardial function associated with viral infection.

Recently, the generation of transgenic knockout mice that lack individual components of the immune system has allowed molecular dissection of immune mechanisms that are important in the pathogenesis of viral myocarditis. In this
issue of Circulation Research, Opavsky et al17 have examined the phenotype of CVB3 infection in mice that lack CD4, CD8, both CD4 and CD8, and the T-cell receptor β chain (TCRβ) in a strain of mice that is susceptible to CVB3 myocarditis. This was accomplished by backcrossing knock-out mice into the susceptible A/J strain of mice that have an H-2k major histocompatibility complex (MHC) haplotype. They found that in the background of a normally robust myocarditic response, knockout of CD4+ lymphocytes caused a small but significant decrease in the inflammatory infiltrate at 14 days after infection. The effect at 7 and 28 days was not statistically significant. There was not a significant change in mortality or cardiac viral titers in the CD4 knockout mice. CVB3 infection of A/J mice that lack CD8+ T cells did not have a significant change in inflammatory infiltrate, mortality, or viral titers when compared with control infected mice.

The most striking finding in the study by Opavsky et al17 was the marked decrease in mortality and inflammatory infiltrate in mice that lacked both CD4+ and CD8+ T cells and the decrease in mortality in TCRβ-deficient mice. In the CD4 and CD8 double knockout mice, there was not a significant change in viral titers, but there was a marked decrease in myocardial TNF-α mRNA 4 days after infection. These experiments demonstrate that in A/J mice, the absence of both CD4+ and CD8+ T-cell subpopulations had a significant beneficial effect after CVB3 infection. However, the absence of CD4+ or CD8+ T cells alone had minimal effect on mortality or myocardial inflammatory infiltration.

Results from previous experiments by Henke et al18 in gene-targeted knockout mice were somewhat different than those reported by Opavsky et al.17 Henke et al18 showed that CVB3 infection of mice that lacked CD4+ lymphocytes had a marked increase in myocardial inflammation, a decrease in cardiac viral titer, and a decrease in mortality compared with control mice. In β2 microglobulin (β2M) knockout mice that lacked CD8+ T cells, but have intact CD4+ T cells, there was no significant change in inflammatory infiltrate in CVB3-infected mice. However, antibody-mediated depletion of CD8+ T lymphocytes in the CD4 knockout mice led to improved mortality and markedly decreased inflammation when compared with CD4 knockout alone. This occurred despite higher viral titers in the CD8 antibody-treated mice.

Comparing the results of Opavsky et al17 to those of Henke et al18 highlights the importance of carefully considering the genetic background in experiments that evaluate the immunopathogenesis of coxsackieviral myocarditis. Wild-type mice with an H-2k MHC haplotype, such as SJ129, frequently used for gene targeting, or C57BL/6, develop little myocarditis after infection with CVB3. However, wild-type mice with the H-2d MHC haplotype, such as A/J mice, develop a potent myocarditis after CVB3 infection.19 Gene-targeted knockout of CD4+ T cells in H-2d mice with a low susceptibility to myocarditis results in a robust CD8-dependent inflammatory response after CVB3 infection, whereas in myocarditic H-2d mice, knockout of CD4+ or CD8+ T cells alone is not sufficient to markedly affect cellular infiltration and mortality. However, the absence of both CD4+ and CD8+ T cells markedly decreases the cellular inflammation and mortality caused by CVB3 infection of both H-2d SJ129 CD4 knockout mice and wild-type H-2d A/J mice.

These findings in genetically deficient CD4 and CD8 mice support previous data that indicated that the cellular inflammatory infiltrate after viral infection in susceptible strains of mice contributes significantly to the mortality and myocardial injury that is associated with coxsackieviral infection. However, the balance between the beneficial and detrimental effects of the immune response in humans is less clear. A multicenter trial of immunosuppressive therapy in patients with myocarditis failed to show a beneficial effect in patients who received immunosuppressive agent(s). The finding that cardiac disease was less severe in patients with a more robust inflammatory response emphasizes the critical nature of the balance between the beneficial and detrimental effects of the immune response.20 Thus, it appears that the immune response needs to be balanced to be optimally effective against infection with CVB3. It must be effective in the destruction of virally infected cells but specific in the attack of only those cells. Imbalance may lead to either overwhelming virus-induced myocardial injury or predominantly immunemediated tissue damage. A better molecular understanding of both the direct effect of viral infection on cardiac myocytes and the balance of beneficial and detrimental effects of the immune response will ultimately provide insight into the mechanisms by which viral infections cause cardiomyopathy in humans.

References


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