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Anti-TNF immunotherapy and tuberculosis reactivation: another mechanism revealed

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Commentary

Anti-TNF immunotherapy has revolutionized the treatment of some inflammatory diseases, such as RA. However, a major concern is that patients receiving this therapy have an increased risk of fungal and bacterial infection, particularly of reactivating latent tuberculosis (TB). In this issue of the *JCI*, in an effort to understand how anti-TNF immunotherapy affects host mechanisms required to control TB, Bruns and colleagues examined the effects of the anti-TNF therapeutic infliximab on *Mycobacterium tuberculosis*–specific human lymphocytes (see the related article beginning on page 1167). The authors report that a granulysin-expressing $CD45RA^+$ subset of effector memory $CD8^+$ T cells that contributes to the killing of intracellular *M. tuberculosis* is depleted in vivo by infliximab in patients with RA, and that these cells are susceptible to complement-mediated lysis in the presence of infliximab in vitro. The study provides insight into host defense mechanisms that act to control TB infection and how they are affected during anti-TNF immunotherapy for autoimmune disease.

bearing multiple myeloma xenografts (7). The multiple myeloma tumors that were treated with R3Mab overexpress wild-type FGFR3. It was convincingly demonstrated that the antitumor effect of R3Mab treatment of tumor cells bearing high amounts of wild-type FGFR3 on their cell surface was mediated in part by Ab-dependent cell-mediated cytotoxicity (ADCC). By contrast, ADCC did not seem to play a role in the antitumor effect of R3Mab on human bladder cancer cells, which express at least a 5-fold lower level of wild-type or mutant FGFR3 compared with the multiple myeloma cells studied.

Finally, since similar gain-of-function mutations in FGFR3 cause skeletal dysplasias and other developmental disorders, it will be important in future studies to examine whether R3Mab treatment may prevent or attenuate developmental disorders caused by FGFR3-activating mutations in neonatal murine models of these diseases (10–12).

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1. Eswarakumar, V.P., Lax, I., and Schlessinger, J. 2005. Cellular signaling by fibroblast growth factor receptors. *Cytokine Growth Factor Rev.* **16**:139–149.
2. Dailey, L., Ambrosetti, D., Mansukhani, A., and Basilico, C. 2005. Mechanisms underlying differential responses to FGF signaling. *Cytokine Growth Factor Rev.* **16**:233–247.
3. Chesi, M., et al. 1997. Frequent translocation t(4;14)(p16.3;q32.3) in multiple myeloma is associated with increased expression and activating mutations of fibroblast growth factor receptor 3. *Nat. Genet.* **16**:260–264.
4. Knowles, M.A. 2008. Novel therapeutic targets in bladder cancer: mutation and expression of FGF receptors. *Future Oncol.* **4**:71–83.
5. Trudel, S., et al. 2004. CHIR-258, a novel, multitargeted tyrosine kinase inhibitor for the potential treatment of t(4;14) multiple myeloma. *Blood.* **105**:2941–2948.
6. Grand, E.K., Chase, A.J., Heath, C., Rahemtulla, A., and Cross, N.C. 2004. Targeting FGFR3 in multiple myeloma: inhibition of t(4;14)-positive cells by SU5402 and PD173074. *Leukemia.* **18**:962–966.
7. Qing, J., et al. 2009. Antibody-based targeting of FGFR3 in bladder carcinoma and t(4;14)-positive multiple myeloma in mice. *J. Clin. Invest.* **119**:1216–1229.
8. Plotnikov, A.N., Schlessinger, J., Hubbard, S.R., and Mohammadi, M. 1999. Structural basis for FGF receptor dimerization and activation. *Cell.* **98**:641–650.
9. Schlessinger, J., et al. 2000. Crystal structure of a ternary FGF-FGFR-heparin complex reveals a dual role for heparin in FGFR binding and dimerization. *Mol. Cell.* **6**:743–750.
10. Chen, L., et al. 1999. Gly369Cys mutation in mouse FGFR3 causes achondroplasia by affecting both chondrogenesis and osteogenesis. *J. Clin. Invest.* **104**:1517–1525.
11. Twigg, S.R., et al. 2009. Skeletal analysis of the Fgfr3(P244R) mouse, a genetic model for the Muenke craniosynostosis syndrome. *Dev. Dyn.* **238**:331–342.
12. Pannier, S., et al. 2009. Activating Fgfr3 Y367C mutation causes hearing loss and inner ear defect in a mouse model of chondrodysplasia. *Biochim. Biophys. Acta.* **2**:140–147.

Anti-TNF immunotherapy and tuberculosis reactivation: another mechanism revealed

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Anti-TNF immunotherapy has revolutionized the treatment of some inflammatory diseases, such as RA. However, a major concern is that patients receiving this therapy have an increased risk of fungal and bacterial infection, particularly of reactivating latent tuberculosis (TB). In this issue of the *JCI*, in an effort to understand how anti-TNF immunotherapy affects host mechanisms required to control TB, Bruns and colleagues examined the effects of the anti-TNF therapeutic infliximab on *Mycobacterium tuberculosis*–specific human lymphocytes (see the related article beginning on page 1167). The authors report that a granulysin-expressing CD45RA$^+$ subset of effector memory CD8$^+$ T cells that contributes to the killing of intracellular *M. tuberculosis* is depleted in vivo by infliximab in patients with RA, and that these cells are susceptible to complement-mediated lysis in the presence of infliximab in vitro. The study provides insight into host defense mechanisms that act to control TB infection and how they are affected during anti-TNF immunotherapy for autoimmune disease.

TNF is a pleiotropic cytokine that plays a critical but incompletely understood role in immunity to *Mycobacterium tuberculosis* and other intracellular bacterial and fungal pathogens. TNF is not only essential for immune control of tuberculosis (TB), it has also been implicated in the immunopathology of the disease (1). While considerable knowledge of the actions of TNF in immunity to *M. tuberculosis* has been gained from studies in animal models, we have also gained a deeper understanding of TNF's contributions to the control of TB in humans through the use of TNF-neutralizing drugs for certain chronic inflammatory diseases. While these agents are highly efficacious for the treatment of RA, ankylosing spondylitis, psoriatic arthritis, and Crohn disease, they also promote reactivation (and possibly acquisition) of intracellular pathogens, including *M. tuberculosis*, resulting in potentially life-threatening infections. Since the early reports of TB in patients treated with infliximab, an anti-TNF monoclonal antibody, described abnormalities in granulomas (2), much of the work to elucidate the mechanisms of TB reactivation has focused on the roles of TNF in the formation and maintenance of these structures. Granulomas are thought to contribute to control of intramacro-

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Nonstandard abbreviations used: CDC, complement-dependent cytotoxicity; TB, tuberculosis; T_{EMRA}, CD45RA$^+$ effector memory T (cell).

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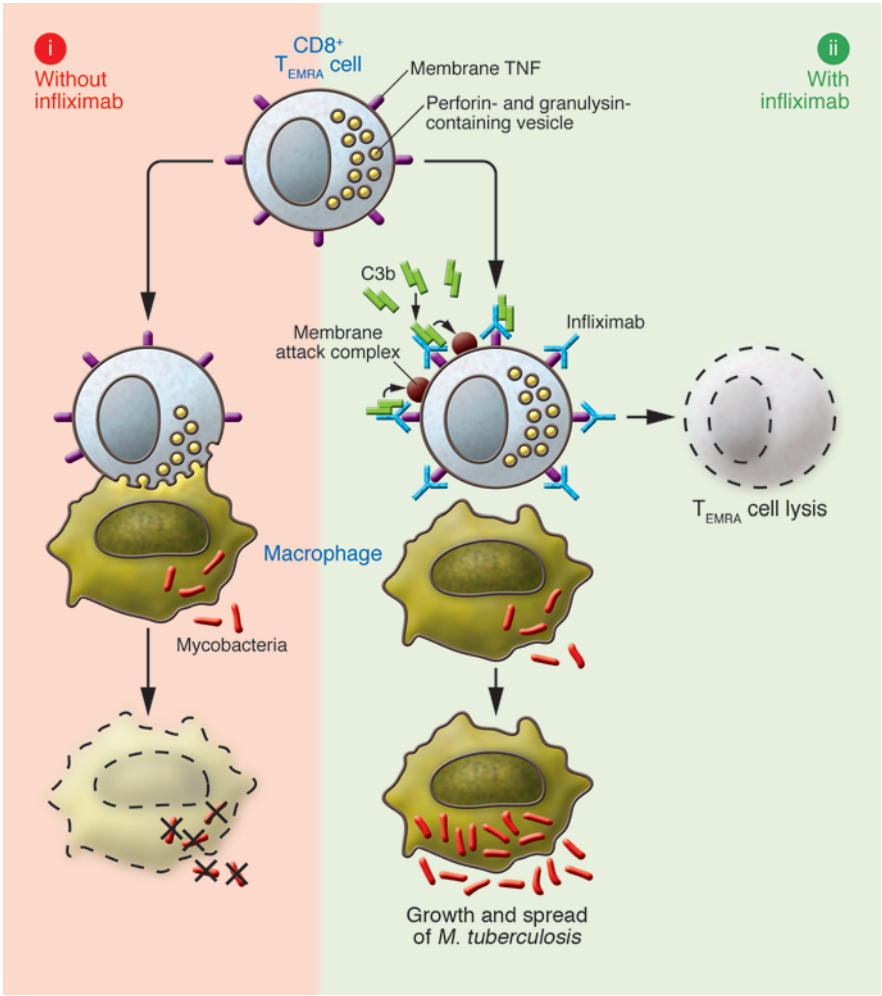

Figure 1
Effect of TNF neutralization with infliximab on the antimycobacterial action of CD8+ T_{EMRA} cells. In the absence of the TNF-neutralizing drug infliximab (i), cytoxic T_{EMRA} cells are present and release their granules containing perforin and granulysin, resulting in the death of *M. tuberculosis*–infected macrophages and intracellular and extracellular mycobacteria. In this issue of the *JCI*, Bruns et al. (3) report that in the presence of infliximab (ii), membrane TNF on T_{EMRA} cells is bound by the antibody, and CDC ensues. The depletion of T_{EMRA} cells results in suboptimal control of mycobacterial growth, leading to the potential spread of *M. tuberculosis* infection.

phage pathogens by providing a local environment in which APCs and lymphocytes interact to suppress progressive growth of the pathogen. In the current issue of the *JCI*, Bruns et al. describe an additional mechanism for susceptibility to progressive TB in individuals treated with infliximab (3). Using PBMCs from tuberculin skin test–positive subjects, Bruns et al. identified granulysin-rich $CD8^+CD45RA^+CCR7^-$ effector memory T cells ($CD8^+$ T_{EMRA} cells) as a major subset of antimycobacterial effector cells and found that T_{EMRA} cells are selectively depleted by infliximab therapy in patients with RA. Here we discuss the impact of these findings as they relate to the existing knowledge of $CD8^+$ T cells and TNF in immunity to *M. tuberculosis*.

$CD8^+$ T cells and immunity to *M. tuberculosis*

Cell-mediated immunity is critical for control of *M. tuberculosis* infection, and it has long been acknowledged that $CD4^+$ T cells are important mediators of immunity to *M. tuberculosis*. More recently, *M. tuberculosis*–specific $CD8^+$ T cells have been increasingly recognized, but their significance remains incompletely understood, especially in humans. $CD8^+$ T cells are thought to limit mycobacterial growth directly through the killing of infected cells (4, 5), as well as indirectly through secretion of cytokines that promote activation of macrophages and chemokines that coordinate cell recruitment. Mice deficient in MHC class I molecules, and thus deficient in $CD8^+$ T cells due to the absence of positive selection, have higher burdens of *M. tuberculosis* in the lungs and a modest decrease in survival (6). This is in contrast to $CD4^+$ T cell–deficient mice, which display more rapidly progressive bacterial growth and a sharp decline in survival (6). These studies and others imply that $CD8^+$ T cells are less crucial during the acute phase of *M. tuberculosis* infection, but are indispensable during the chronic phase of infection, and therefore may help prevent reactivation of TB (7, 8). When evaluating the contribution of $CD8^+$ T cells to *M. tuberculosis* immunity, it is important to note that mice lack granulysin, a cytolytic granule protein that contributes to killing of *M. tuberculosis* by human $CD8^+$ T cells (4). Therefore, murine studies may undervalue the importance of the antimycobacterial properties of $CD8^+$ T cell subsets compared with their roles in human immunity. Human studies are more limited in number, but in vitro experiments provide evidence that $CD8^+$ T cells can control *M. tuberculosis* through killing of infected macrophages and subsequent death of the bacteria, as well as by direct killing of mycobacteria by secreted granulysin (4, 5, 9). Bruns et al. (3) found in their assays that T_{EMRA} cells stain for granulysin with high frequency and display the highest levels of both cytotoxicity and antimycobacterial activity compared with other T cell subsets.

TNF blockade and progression of *M. tuberculosis*

The authors go beyond defining T_{EMRA} cells as potentially important antimycobacterial effector cells, to address the consequences of TNF blockade on these cells and on mycobacterial growth in cultured cells (3). In doing so, they reveal what we believe to be a novel potential mechanism for TNF neutralization to promote progression of TB. Previous studies in mice and in zebrafish embryos have revealed variable increases in bacillary load in the absence of TNF, ranging from small changes to multiple orders of magnitude (10–13). These observations indicated that TNF contributes directly to control of mycobacteria in granuloma macrophages (10) and may also function indirectly to suppress mycobacterial growth by modulating formation and/or maintenance

of granulomas. Additional evidence suggests that TNF blockade may impair mycobacterial control as a result of effects on T lymphocytes. TNF blockade has previously been shown to induce production of Tregs via TGF-β in patients with RA (14, 15), and in mice infected with *M. tuberculosis*, depletion of Tregs leads to improved control of mycobacterial growth (16). Bruns et al. provide evidence that TNF blockade may also promote reactivation of TB through selective reduction of T_{EMRA} cells (3). As a mechanism for their observation that T_{EMRA} cells in peripheral blood are depleted following infliximab therapy, the authors demonstrate that T_{EMRA} cells from RA patients possess high levels of membrane-bound (presumably transmembrane) TNF compared with healthy controls and are susceptible to in vitro complement-mediated lysis in the presence of infliximab. Additionally, PBMCs from patients receiving infliximab therapy displayed diminished capacity to limit mycobacterial growth in culture, an impairment that was rescued by the addition of autologous $CD8^+$ T_{EMRA} cells, to a slightly lesser extent by $CD8^+CCR7^-CD45RA^-$ effector memory T cells, but not by circulating central memory $CD8^+$ T cells.

Implications for future study

As with any provocative results, the findings reported by Bruns et al. (3) raise several questions, with implications for understanding immunity to TB and with practical application to clinical medicine. First, do infliximab and etanercept differ in their ability to deplete T_{EMRA} cells? Infliximab, a monoclonal antibody to TNF, and etanercept, a soluble TNF receptor, are both TNF-neutralizing agents used in the treatment of RA and other inflammatory conditions. For unclear reasons, infliximab causes significantly more infectious complications, including reactivation of *M. tuberculosis*, than does etanercept; this has been attributed by some to the differential ability of these agents to bind membrane TNF. Several studies have found that apoptosis ensues upon binding of infliximab to membrane TNF on T cells (17), monocytes (18), and cells transfected to express membrane TNF (19, 20), which may lead to a reduction in the number of antimycobacterial effector cells and/or to dissolution of granulomas. In contrast, etanercept does not cause apoptosis of cells that express membrane TNF (17, 19, 20). Additionally, cell lines transfected with membrane TNF have been used to evaluate complement-dependent cytotoxicity (CDC) and antibody-dependent, cell-mediated cytotoxicity (ADCC); while both etanercept and infliximab lead to ADCC, only infliximab induces CDC (20, 21). Consistent with these results, Bruns et al. (3) revealed that T_{EMRA} cells are susceptible to CDC in the presence of infliximab (Figure 1). If etanercept does not cause CDC of T_{EMRA} cells, this outcome would suggest that the differential risk of reactivation of TB with these agents may be caused, at least in part, by relative differences in depletion of T_{EMRA} cells. Likewise, if etanercept does not deplete T_{EMRA} cells to the same extent as does infliximab, this could at least partially explain the therapeutic differences observed with these 2 drugs. While both drugs are equally efficacious for the treatment of RA, infliximab is superior to etanercept in the treatment of other chronic inflammatory diseases, especially Crohn disease (22). If selective reduction of T_{EMRA} cells does not occur following etanercept therapy, this finding could provide insight into differences in the immunopathology of these inflammatory conditions. In contrast, if etanercept does deplete T_{EMRA} cells to the same extent as does infliximab, it would weaken the case that T_{EMRA} cells are an essential element of protective immunity against TB.

Second, how long does infliximab-mediated depletion of T_{EMRA} cells persist? The frequency of progressive TB in people treated with infliximab is highest in the first 90 days after initiating the therapy (23), but the studies by Bruns et al. were limited to a single time point 2 weeks after initiating therapy (3). Additional studies of the duration of depletion of T_{EMRA} cells after anti-TNF therapy should shed further light on the roles of these cells in protection against TB.

Third, what are the antigens recognized by T_{EMRA} cells, and what determines differentiation of $CD8^+$ effector cells into T_{EMRA} cells rather than $CD45RA^-$ effector memory T cells? Answers to these questions could provide guidance in the design of improved TB vaccines by delineating optimal antigens and adjuvants and might also provide insight into the mechanisms of differential susceptibility and resistance to TB in populations and in individuals. In addition, they may identify Ag-specific T_{EMRA} cells as potential surrogate markers of vaccine efficacy and thereby provide improved predictive information in the design and evaluation of clinical trials of novel TB vaccines.

Finally, while the findings reported by Bruns et al. clearly demonstrate that T_{EMRA} cells are targets of the anti-TNF action of infliximab (3), they do not provide proof that TNF has an essential function in the development, maintenance, or effector functions of this interesting subset of $CD8^+$ T cells. If membrane TNF is simply a bystander, then future development of agents that block TNF activities without depleting T_{EMRA} cells may provide for safer therapy of chronic inflammatory diseases.

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1. Mohan, V.P., et al. 2001. Effects of tumor necrosis factor alpha on host immune response in chronic persistent tuberculosis: possible role for limiting pathology. *Infect. Immun.* **69**:1847–1855.
2. Keane, J., et al. 2001. Tuberculosis associated with infliximab, a tumor necrosis factor alpha-neutralizing agent. *N. Engl. J. Med.* **345**:1098–1104.
3. Bruns, H., et al. 2009. Anti-TNF immunotherapy reduces $CD8^+$ T cell–mediated antimicrobial activity against *Mycobacterium tuberculosis* in humans. *J. Clin. Invest.* **119**:1167–1177.
4. Stenger, S., et al. 1998. An antimicrobial activity of cytolytic T cells mediated by granulysin. *Science.* **282**:121–125.
5. Stenger, S., et al. 1997. Differential effects of cytolytic T cell subsets on intracellular infection. *Science.* **276**:1684–1687.
6. Mogues, T., Goodrich, M.E., Ryan, L., LaCourse, R., and North, R.J. 2001. The relative importance of T cell subsets in immunity and immunopathology of airborne Mycobacterium tuberculosis infection in mice. *J. Exp. Med.* **193**:271–280.
7. Ordway, D., et al. 2007. XCL1 (lymphotactin) chemokine produced by activated CD8 T cells during the chronic stage of infection with Mycobacterium tuberculosis negatively affects production of IFN-gamma by CD4 T cells and participates in granuloma stability. *J. Leukoc. Biol.* **82**:1221–1229.
8. van Pinxteren, L.A., Cassidy, J.P., Smedegaard, B.H., Agger, E.M., and Andersen, P. 2000. Control of latent Mycobacterium tuberculosis infection is dependent on CD8 T cells. *Eur. J. Immunol.* **30**:3689–3698.
9. Carranza, C., et al. 2006. Mycobacterium tuberculosis growth control by lung macrophages and CD8 cells from patient contacts. *Am. J. Respir. Crit. Care Med.* **173**:238–245.
10. Clay, H., Volkman, H.E., and Ramakrishnan, L. 2008. Tumor necrosis factor signaling mediates resistance to mycobacteria by inhibiting bacterial growth and macrophage death. *Immunity.* **29**:283–294.
11. Flynn, J.L., et al. 1995. Tumor necrosis factor-alpha is required in the protective immune response against Mycobacterium tuberculosis in mice. *Immunity.* **2**:561–572.
12. Kindler, V., Sappino, A.P., Grau, G.E., Piguet, P.F., and Vassalli, P. 1989. The inducing role of tumor necrosis factor in the development of bactericidal granulomas during BCG infection. *Cell.* **56**:731–740.
13. Zganiacz, A., et al. 2004. TNF-alpha is a critical negative regulator of type 1 immune activation dur-

ing intracellular bacterial infection. *J. Clin. Invest.* **113**:401–413.
14. Ehrenstein, M.R., et al. 2004. Compromised function of regulatory T cells in rheumatoid arthritis and reversal by anti-TNFalpha therapy. *J. Exp. Med.* **200**:277–285.
15. Nadkarni, S., Mauri, C., and Ehrenstein, M.R. 2007. Anti-TNF-alpha therapy induces a distinct regulatory T cell population in patients with rheumatoid arthritis via TGF-beta. *J. Exp. Med.* **204**:33–39.
16. Scott-Browne, J.P., et al. 2007. Expansion and function of Foxp3-expressing T regulatory cells during tuberculosis. *J. Exp. Med.* **204**:2159–2169.
17. Van den Brande, J.M., et al. 2003. Infliximab but not etanercept induces apoptosis in lamina propria T-lymphocytes from patients with Crohn's disease. *Gastroenterology.* **124**:1774–1785.
18. Lugering, A., et al. 2001. Infliximab induces apoptosis in monocytes from patients with chronic active Crohn's disease by using a caspase-dependent pathway. *Gastroenterology.* **121**:1145–1157.
19. Mitoma, H., et al. 2005. Infliximab induces potent anti-inflammatory responses by outside-to-inside signals through transmembrane TNF-alpha. *Gastroenterology.* **128**:376–392.
20. Mitoma, H., et al. 2008. Mechanisms for cytotoxic effects of anti-tumor necrosis factor agents on transmembrane tumor necrosis factor alpha-expressing cells: comparison among infliximab, etanercept, and adalimumab. *Arthritis Rheum.* **58**:1248–1257.
21. Scallon, B.J., Moore, M.A., Trinh, H., Knight, D.M., and Ghrayeb, J. 1995. Chimeric anti-TNF-alpha monoclonal antibody cA2 binds recombinant transmembrane TNF-alpha and activates immune effector functions. *Cytokine.* **7**:251–259.
22. Sandborn, W.J., et al. 2001. Etanercept for active Crohn's disease: a randomized, double-blind, placebo-controlled trial. *Gastroenterology.* **121**:1088–1094.
23. Dinarello, C.A. 2005. Differences between anti-tumor necrosis factor-alpha monoclonal antibodies and soluble TNF receptors in host defense impairment. *J. Rheumatol. Suppl.* **74**:40–47.

CaMKII and a failing strategy for growth in heart

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Patients with systolic left ventricular dysfunction die progressively from congestive heart failure or die suddenly from cardiac arrhythmias. Myocardial hypertrophy is an early event in most forms of heart failure, but the majority of patients with myocardial hypertrophy do not develop heart failure. Developing improved therapies for targeting the cell signaling pathways that enable this deadly transition from early myocardial insult to heart failure and sudden death is a key goal for improving public health. In this issue of the *JCI*, Ling and colleagues provide new evidence that activation of the multifunctional Ca^{2+}/calmodulin–dependent kinase IIδ is a decisive step on the path to heart failure in mice (see the related article beginning on page 1230).

The heart failure epidemic

Heart failure is a leading cause of death in the developed world, and the incidence of heart failure continues to rise despite impressive improvements in treating coronary artery disease and hypertension. Heart failure patients encounter 2 main problems: reduced cardiac pumping that is inadequate to meet metabolic demands, and electrical instability that causes arrhythmias and sudden death. Heart failure is a clinical syndrome, but most heart failure in the developed world is caused by reduced left ventricular systolic function as a result of hypertension and myocardial infarction. Antihypertensive medications have resulted in a reduction in the number of patients with myocardial hypertrophy (1), and improved treatment of myocardial infarction patients has drastically reduced the acute and 5-year mortality of this common form of structural heart disease (2). Unfortunately, the incidence of heart failure has increased, despite reducing the burden of hypertension and the lethality of myocardial infarction (2). Improved understanding of myocardial biology favoring conversion of early forms of structural heart disease into the advanced syndrome of heart failure and sudden death is needed to devise more effective approaches to preventing and treating heart failure.

Why is CaMKII important in myocardial disease?

Many signaling molecules are implicated in structural heart disease. However, few, if any, molecules appear to be as central to pathological response mechanisms in heart as Ca^{2+}/calmodulin–dependent kinase II (CaMKII). CaMKII expression and activity are increased in failing human myocardium and in many animal models of cardiac hypertrophy and heart failure. CaMKII overexpression in mouse myocardium causes hypertrophy, failure, and sudden death (3), while CaMKII inhibition suppresses these phenotypes (4, 5). Pathological stress induces a cardiomyocyte milieu of dysregulated and prolonged cytoplasmic Ca^{2+} transients (6) and increased oxidative stress (7). CaMKII is an ideal nodal molecule for transducing Ca^{2+} and redox signals — fundamental upstream signals common to most forms of structural heart disease — into downstream events, such as apoptosis (8), hypertrophy (3), and proarrhythmic electrical remodeling (3, 4), that lead to the clinical phenotypes of congestive heart failure and sudden death (Figure 1A). Finally, CaMKII itself amplifies the disruption of intracellular Ca^{2+} homeostasis in heart failure by increasing the opening probability of voltage-gated Ca^{2+} channels (9) and ryanodine receptors, the latter being the major mediators of Ca^{2+}-induced Ca^{2+} release from intracellular sarcoplasmic reticulum Ca^{2+} stores in cardiac myocytes (3).

CaMKII is initially activated by binding to calcified calmodulin (Ca^{2+}/CaM). Ca^{2+}/CaM binding reorders the structure of the CaMKII molecule by disinhibiting the catalytic domain and exposing the regulatory domain (Figure 1B). The CaMKII regulatory domain is the target for activity-sustaining modifications that bring about the transition of CaMKII into a Ca^{2+}/CaM–autonomous enzyme. Ca^{2+}/CaM–activated CaMKII monomers are susceptible to autophosphorylation (10) at threonine 286/287 (the specific number is isoform dependent) and oxidation of paired methionines 281 and 282 (8). CaMKII autophosphorylation and oxidation prevent

Conflict of interest: Mark E. Anderson is named as the inventor on patents issued and pending regarding the treatment of heart failure and arrhythmias by CaMKII inhibition.

Nonstandard abbreviations used: Ca^{2+}/CaM, calcified calmodulin; CaMKII, Ca^{2+}/calmodulin–dependent kinase II; HDAC, class II histone deacetylase; MEF2, myocyte enhancer factor 2.

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