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Ataxia-Telangiectasia Mutated Kinase: A Potential New Target for Suppressing Inflammation in Heart Failure?

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Ataxia-telangiectasia mutated (ATM) kinase, the mutation of which causes the autosomal recessive disease ataxia-telangiectasia, plays an essential role in the maintenance of genome stability (reviewed in ref. 1). ATM (a serine/threonine protein kinase) senses DNA double-strand breaks and phosphorylates several key proteins to initiate the DNA damage response, leading to cell cycle arrest, DNA repair, or apoptosis.[2] In fact, ATM is one of the master regulators of the cellular response to radiation-induced DNA damage and a key determinant of radiosensitivity. DNA damage leads to activation of ATM kinase activity and phosphorylation of a number of downstream targets such as p53, CHK2, and KAP-1.[3,4] This activation triggers cell cycle checkpoints, arrest, and delays in the G_1, S, and G_2 phases of the cell cycle and enables DNA repair of double-stranded breaks both by homologous recombination and by non-homologous end joining. Hence, fibroblasts and tumor cells are radiosensitized to x-ray radiation therapy in culture by pharmacological ATM inhibition, or by ATM mutation and deletion.[5] ATM deficiency has been shown to sensitize cells to inhibition of poly (ADP-ribose) polymerase (PARP), an enzyme involved in DNA repair and apoptosis. Conversely, abnormally active ATM also impairs DNA repair by homologous recombination and thereby sensitizes cells to PARP inhibition. Thus, timely activation and inactivation of ATM are both necessary for efficient repair, and any ATM perturbation could inhibit the ability of cells to resist DNA damage.[6] Clinically, it has been shown that cells isolated from patients with ataxia telangiectasia lacking functional ATM are sensitive to ionizing radiation.[7] The chemotherapy drug doxorubicin also activates ATM through the production of superoxide radicals and induces apoptosis via p53.[8] However, the role of ATM in myocardial infarction (MI) has not been studied as extensively as cancer although ATM-dependent signaling has been suggested to play a role in the development of atherosclerotic vascular disease.[9]

Heart failure usually leads to increased chamber diameter which results in increased loading capacity of the heart represented by increased left ventricular end-systolic volume (LVESV) and left ventricular end-diastolic volume (LVEDV). Increased LVESV is suggested as one of the major determinants of survival, post-MI.[10] Low-level but progressive loss of myocytes in the chronically overloaded heart is believed to contribute to cardiac remodeling and contractile failure (reviewed in ref. 11). Apoptosis in the heart following MI can be triggered by activation of G-protein coupled receptors (GPCRs), cytokines, and increased generation of ROS. Several kinases including ASK1 (apoptosis signal-regulating kinase 1), p38MAPK, JNK (c-Jun N-terminal kinase), CaMKII as well as protein kinase C-dependent transcriptional upregulation of the pro-apoptotic protein NIX (also known as BNIP3L) target mitochondria.[12] CaMKII is potentially the convergence of pro-apoptotic signaling because it is activated by both Ca^{2+} and regulated production of NADPH oxidase (NOX)-derived ROS, downstream of angiotensin II-induced stimulation of GPCRs.[13] Apoptotic cell death is counteracted by pro-survival pathways, such as activation of Akt and proto-oncogene serine-threonine protein kinase (PIM1) and inactivation of glycogen synthase kinase 3ß (GSK3β).[14]

Programmed necrosis is a different type of cell death that has also been suggested to be important in heart disease.[15] Necrosis is accompanied by early loss of plasma membrane and organelle integrity and striking inflammation. Inflammation can contribute to extracellular matrix remodeling and development of contractile failure. An important feature of the programmed necrosis is opening of the mitochondrial permeability transition pore (MPTP) in response to mitochondrial Ca^{2+} and perhaps oxidative stress. Opening of MPTP causes collapse of mitochondrial membrane potential and ATP production and triggers necrosis. It has also been shown there is crosstalk between the apoptotic and necrotic

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pathways, facilitated by Bcl-2 family proteins and the MPTP. Bax and Bak are known to play a primary role in activating apoptosis in response to myocardial ischemia and reperfusion, and Bax/Bak double knockout mice exhibit reduced infarcts compared with wild type mice (reviewed in ref. 16). However, Bax/Bak/cyclophilin D triple knockout mice do not show further reduction in infarct size compared with the Bax/Bak double knockout mice. In addition, cells and mitochondria lacking Bax and Bak are resistant to mPTP opening and necrosis, suggesting that Bax and Bak play distinct roles in regulating both apoptosis and necrosis.

MI also triggers an intense inflammatory response, which is essential for cardiac repair as well as post-infarction remodeling and heart failure.[17] Neutrophils recruited to the infarcted area remove dead cells and matrix debris by phagocytosis, while preparing the area for scar formation. Attraction of inflammatory cells could be stimulated by programmed myocyte necrosis within the heart, which may release damage-associated molecular patterns (DAMPs) from the cytosol and provoke inflammatory response by activation of the innate immune system.[18,19] The stressed myocytes signal to fibroblasts and other cells within the matrix through release of factors such as connective tissue growth factor (CTGF) and transforming growth factor β (TGFβ).[20] Members of the TGF-ß family are critically involved in suppression of inflammation and activation of a pro-fibrotic program.[17]

As stated before, there is little information on the role of ATM in relation to post-MI remodeling, inflammation and apoptosis in the heart. Previous work from Singh and colleagues showed that ATM deficiency attenuates LV dysfunction and dilatation 7 days post-MI.[21] In addition, they provided evidence that ATM deficiency resulted in increased cardiac fibrosis and expression of α-smooth muscle actin (α-SMA, a marker formyofibroblasts) in the infarct region 7 days post-MI.[21] In the paper by Daniel et al,[22] the authors have further studied the effects of ATM deficiency on the inflammatory response, and activation of survival signaling molecules including Akt and GSK-3ß in the heart following acute MI. Using ATM heterozygous knockout (hKO) and corresponding wild-type mice subjected to MI by occlusion of coronary artery, these authors studied cardiac function, infarct size neutrophil infiltration, macrophages, apoptosis, fibrosis and survival signaling. The results showed that MI increased neutrophil infiltration in the infarct regions of LV in both genotypes on day 1 and 3 post-MI when compared with their respective sham groups. Interestingly, the number of neutrophils was significantly lower in the infarct and non-infarct LV regions of hKO-MI when compared with WT-MI 1 day post-MI. Similarly the number of macrophages was significantly lower in the infarct LV region of hKO-MI versus WT-MI 1 day post-MI. The number of macrophages was not significantly different between the 2 genotypes 3 days post-MI although

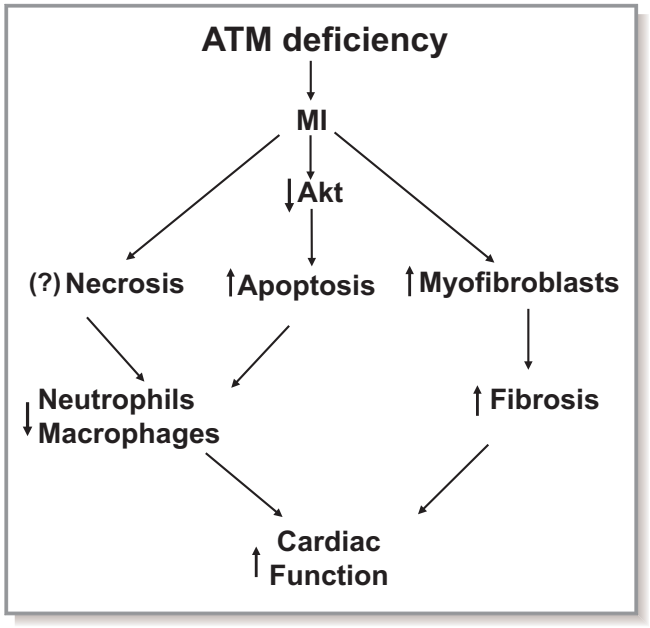

Figure. Schematic showing how ATM deficiency may influence heart function early post-MI. ATM deficiency decreases activation of anti-apoptotic signaling kinase, p-Akt, and increases activation of pro-apoptotic signaling, p-GSK-3β, resulting in increased apoptosis. The increased apoptosis may have inhibitory effect on inflammatory response. Although not investigated in this study, necrosis can potentially influence the inflammatory response as well. ATM deficiency also increases myofibroblast activation thereby increasing fibrosis. This early increase in fibrosis and/or decreased inflammatory response may help maintain cardiac function early post-MI. ATM indicates ataxia-telangiectasia mutated; GSK-3 β, glycogen synthase kinase 3ß; MI, myocardial infarction.

they were still higher in number in the infarct LV regions of both genotypes when compared with their respective sham groups. Levels of active TGF-β1 were reduced in the infarct area during ATM deficiency 3 days post-MI. ATM deficiency was associated with increased apoptosis, fibrosis and expression of α-SMA in the heart post-MI (Figure). Moreover, the activation of pro-survival kinase, Akt, was lower while activation of pro-apoptotic kinase, GSK-3β, was higher in ATM deficient hearts 1 day post-MI. The ejection fraction or fractional shortening were not different between the 2 genotypes 3 days post-MI, although LVESV and LVEDV were significantly lower in ATM-deficient hearts at both time points. The better LV function 1 day post-MI during ATM deficiency did not correlate with infarct size which remained unchanged between the 2 genotypes 1 and 3 days post-MI. Overall, despite mixed results, these studies suggest that ATM has the potential to modulate the remodeling processes in the heart post-MI during early phase. However, impact of long-term ATM deficiency in remodeling and healing of the infarcts still remains uncertain at this time.

The authors are to be commended for investigating the potential novel role of ATM deficiency in attenuation of inflammation in post-MI remodeling. During the last few years, pharmaceutical industries and research laboratories have developed a series of small molecules, capable of inhibiting ATM kinase with increasing specificity in cancer cells. One such inhibitor, KU60019 has been shown to be a potent chemo sensitizer in combination with doxorubicin in breast cancer cells.[23] In order to expand the scope of these investigations with the hope of finding their potential use in patients with MI, the novel ATM kinase inhibitors need to be carefully evaluated for their possible remodeling and anti-inflammatory effects in post-MI heart failure.

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None.

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