Therapeutic potential of pharmacological inhibition of programmed apoptosis, necroptosis, and ferroptosis in improving left ventricular function in post-myocardial infarction rats

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Background: Heart failure (HF) is considered the most fatal complicating sequelae after myocardial infarction (MI). Various types of programmed cell deaths (PCDs), including apoptosis, necroptosis, and ferroptosis, have been documented as the pathological progression following MI. Although novel pharmacologically specific cell death inhibitors of apoptosis, necroptosis, and ferroptosis have been shown to exert cardioprotection against acute MI and reperfusion injury, their beneficial effects have never been investigated in a post-MI-induced HF model.

Purpose: To investigate the therapeutic efficacy and potential underlying mechanisms of inhibitors of apoptosis, necroptosis, and ferroptosis in post-MI rats.

Methods: Adult male Wistar rats were assigned to the sham (n = 6) and the MI groups (n = 30) via a permanent LAD occlusion. After 1-week LAD ligation, MI rats were divided into 5 subgroups: 1) vehicle (3% DMSO/day, IP, CON, n = 6), 2) enalapril (10 mg/kg/day, PO, ENA, n = 6), 3) apoptosis inhibitor zVAD-FMK (1 mg/kg/day, IP, zVAD, n = 6), 4) necroptosis inhibitor Necrostatin-1 (1.65 mg/kg/day, IP, Nec-1, n = 6), or 5) ferroptosis inhibitor Ferrostatin-1 (2 mg/kg/day, IP, Fer-1, n = 6) for 4 weeks. Then, echocardiography for LV function was performed, and the hearts were removed to determine pertinent apoptosis, necroptosis, and ferroptosis proteins by immunoblotting assays. Malondialdehyde (MDA) levels for myocardial oxidative stress were quantified.

Results: Post-MI rats exhibited worsening LV dysfunction, as indicated by a markedly reduced %LV ejection fraction (LVEF) (Fig. 1A). Post-MI rats showed higher MDA levels in cardiac tissue, indicating myocardial oxidative stress (Fig. 1B). Furthermore, post-MI rats had elevated expressions of proteins related to apoptosis (Cleaved caspase3/Caspase 3) and necroptosis (pMLKL/MLKL), but not ferroptosis (ACSL4/GAPDH) (Fig. 1C-E). As a standard treatment for MI, enalapril reduced not only myocardial oxidative stress, but also apoptosis and necroptosis activation, thereby enhancing %LVEF after MI (Fig 1A-D). Notably, treatment of post-MI rats with the apoptosis inhibitor zVAD-FMK, necroptosis inhibitor Necrostatin-1, and ferroptosis inhibitor Ferroptosin-1 ameliorated LV dysfunction and myocardial oxidative stress upregulation (Fig A-B). Interestingly, treatment with zVAD, Nec-1, and Fer-1 effectively attenuated both myocardial apoptosis and necroptosis activation after MI (Fig 1C-D).

Conclusion: Post-MI increased myocardial oxidative stress and activated multiple PCDs, including apoptosis and necroptosis but not ferroptosis, thus leading to LV dysfunction. Apoptosis and necroptosis inhibitors effectively mitigated oxidative stress and myocardial death, resulting in improved LV function in post-MI rats. These findings highlight the therapeutic potential of inhibiting apoptosis and necroptosis as promising pharmacological interventions to improve clinical outcomes for post-MI patients.