CARDIAC FIBROBLASTS SECRETOME PROTECTS CARDIOMYOCYTES FROM SIMULATED ISCHEMIA REPERFUSION INJURY

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INTRODUCTION

BACKGROUND: Cardiac fibroblasts (CF) are the largest cell population of the heart. Previous studies showed their involvement in physiological but also pathological conditions like heart failure and remodeling; however their role during a sequence of ischemia reperfusion has never been studied. Preliminary data obtained in our lab showed that cardiomyocyte viability was modulated by the presence of CF.

HYPOTHESIS: We hypothesized that CF were implicated in the modulation of cardioprotection against ischemia reperfusion injury. We aimed to determine : (1) if this effect is paracrine (2) when this type of action is effective ? (3) What kind of factors that maybe involved in the protection are secreted by CF?

METHODS

Neonatal rat cardiomyocytes (NRC) and CF were isolated from Wistar rats hearts and cultured separately. NRC were submitted to simulated ischemia reperfusion in two different conditions: (1) in the presence of CF placed in insert (2) after addition of conditioned supernatant of CF. Isolated cell cultures or co-cultures were subjected to 24 H normoxia or 3 H of simulated ischemia followed by 21 H of simulated reperfusion. At the end of reperfusion, MTT cell viability assay and troponin I measurement to quantify cell death, were performed. Cytokine array was performed with supernatant of CF after 3 H of ischemia. Data are means ± SEM. Groups comparison were performed by ANOVA.

EXPERIMENT GROUPS

All groups (n=6/group) were done with the same number of NRC (5*10^5 cells/dish) and the same number of CF in inserts (2*10^5 cells/insert).

RESULTS

Figure 1 showed that CF placed in the insert protect NRC during I/R through a paracrine mechanism and that this effect was observed when CF were added before ischemia but not at the time of reperfusion. Data obtained for troponin I measurement were correlated with MTT (data not shown).

Figure 2 showed that SN of CF protected NRC during I/R through a paracrine mechanism and that this action seemed to be effective when SN was added at reperfusion. Three potentially protective factors have been detected in CF secretome : TIMP-1, CXCL-7 and IFN-γ.

CONCLUSION

Our data suggest, for the first time, that CF participates in endogenous cardioprotection against ischemia reperfusion injury. This protection is mediated by paracrine mechanisms and the secretome of CF is more effective when added at reperfusion. Three potentially protective factors have been detected in CF secretome : TIMP-1, CXCL-7 and IFN-γ.