Summary

In this Ph.D. thesis, following aims were addressed: 1) potentially cardioprotective effects of deferiprone on the model of daunorubicin-induced chronic cardiotoxicity in rabbits, 2) the role of apoptotic cell death in the development of anthracycline cardiotoxicity, 3) cardioprotective effects of dexrazoxane against chronic anthracycline cardiotoxicity with a focus on rescue of cardiac myocytes from programmed cell death and oxidative stress, and 4) staging of myocardial changes in the time-course of chronic anthracycline cardiotoxicity development.

First, using the leukemic cell line, deferiprone (1-300 μmol/L) was shown not to blunt the antiproliferative effect of daunorubicin. Instead, at higher concentrations of deferiprone, the augmentation of antiproliferative actions of both agents was observed. However, in the cardioprotective study deferiprone failed to afford significant protection against daunorubicin-induced mortality, cardiac dysfunction, morphological cardiac deteriorations, plasma cardiac troponin T rise as well as myocardial lipoperoxidation. This finding contrasted with previous positive outcomes of *in vitro* studies. Hence, this study changes the current view on deferiprone as a potential cardioprotectant against anthracycline cardiotoxicity. In addition, these results, together with our previous findings, further suggest that the role of iron and its chelation in anthracycline cardiotoxicity is not as trivial as originally believed and/or other mechanisms unrelated to iron-catalyzed ROS production might be involved.

The study on apoptotic cell death using *in vitro* model of anthracycline cardiotoxicity revealed that at the lower (clinically relevant) concentrations of daunorubicin induced this mode of cell death. Furthermore, it has been observed that non-programmed (necrotic) cell death may prevail in the higher daunorubicin concentration (above 3 μmol/l), which is in line with previously reported data. This study revealed also dose dependent increase in the activity of caspases 3 a 9. Hence, these outcomes suggest that intrinsic (mitochondrial) apoptotic pathway is pivotal in daunorubicin-induced programmed cell death in H9c2 cell line. Extrinsic and endoplasmic/sarcoplasmic reticulum pathways seem to not be involved in this process.

Dexrazoxane was capable of fully overcoming premature death of animals, development of systolic dysfunction and plasma cardiac troponins rise. Dexrazoxane was found to rescue the cardiac myocytes from the progressive degeneration and consequently non-programmed cell death. Furthermore, this effective cardioprotection was associated with protection of the left ventricular myocardium from remodeling. For the first time, it has been demonstrated that dexrazoxane is able to protect cardiomyocytes against anthracycline-induced apoptotic cell death. Dexrazoxane was clearly shown to effectively suppress the complex apoptotic signaling triggered by daunorubicin. Hence, this study pointed out on inhibition of apoptosis as a substantial part of cardioprotective action of dexrazoxane against anthracycline cardiotoxicity. Moreover, it was revealed that overcoming of lipoperoxidation need not play a key role in dexrazoxane-afforded cardioprotection. Furthermore, these findings underline a recent call for revisiting the traditional "ROS and iron hypothesis" of pathogenesis of anthracycline cardiotoxicity and mechanisms of dexrazoxane-afforded cardioprotection.

Finally, in this work the dynamic changes in functional, morphological and other parameters in the time-course of anthracycline cardiotoxicity development were for the first time followed. Significant decrease of the left ventricular systolic function together with plasma cardiac troponins rise have been observed, starting by the 8^{th} administration of daunorubicin (cumulative dose 400 mg/m^2). Analyses with individual substrates specific for different matrix metalloproteinases led to analogous outcomes suggesting a trend to MMPs activity elevation with the start of the study, subsequent stabilization and significant MMPs activity rise at the end of the experiment. The principal role is likely to be attributable to activity of MMP-12. In the second part of the experiment, significant elevation of oxidative stress markers was documented. The character of changes of these parameters strongly suggested that these alterations are not cumulative with the maximum being present before the left ventricular systolic dysfunction and morphological deteriorations. These outcomes suggest that oxidative stress might be rather triggering than executive factor in the development of chronic anthracycline cardiotoxicity.