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EFFECT OF RIL-10 ON CATHEPSIN B ACTIVITY INDUCED IN FETAL RAT ALVEOLAR TYPE II CELLS EXPOSED TO HYPEROXIA

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Backgrounds and aims: Hyperoxia induces lung injury through a main biological effect of cell death. Oxygen species generated by hyperoxia can induce lysosomal permeabilization, through which proteolytic enzyme, cathepsin B(CB) is released. We have defined that fetal alveolar type II cell (FATIIC) death was mediated by CB during 65%-hyperoxia and pre-incubation of rIL-10 to FATIICs had beneficial effect on reducing cell death secondary to 65%-hyperoxia. There have never been any trials to investigate the effect of rIL-10 on CB activity in FATIICs exposed to hyperoxia. We speculated that pre-incubation of rIL-10 might inhibit CB activity in FATIICs during 65%-hyperxia.

Methods: FATIICs were isolated on E19(term=22) and exposed to 65%-oxygen for 24 h and 36 h. Cells in room air was used as controls. Cytotoxicity was analyzed by LDH release. Apoptosis was analyzed by TUNEL assay and FACsan. Caspase-3 activity was analyzed by colorimetric assay and western blotting. CB activity was assessed by fluorescence-based assay, western blotting and qRT-PCR. After pre-incubation with rIL-10, CB activity was re-analyzed by the identical methods.

Results: 65%-hyperoxia increased LDH-release significantly in a time-dependent manner compared to controls. Caspase-3 activities were not detected in FTAIICs during 65%-hyperoxia, whereas CB activities were increased greatly during 65%-hyperoxia in a time-dependent manner. After pre-incubation with rIL-10, CB activities were decreased significantly in FATIICs compared to the cells without rIL-10. Similar findings were observed on western blots and qRT-PCR.

Conclusions: Administration of rIL-10 might play a role to reduce FATIIC death secondary to hyperoxia by inhibiting CB activity.