Delivery of low molecular weight antioxidants into rat lungs post exposure to Cl₂ gas

Andreas Bracher¹, Stephen Doran¹, Giuseppe L. Squadrito², John Trombley³, Larry Bowen³ and Sadis Matalon⁴

Departments of Anesthesiology², Environmental Health Sciences³ Schools of Medicine and Public Health, University of Alabama at Birmingham, and, Infectious Diseases Research, Southern Research Institute, Birmingham, AL, USA (SUPPORTED BY: 1U01ES015676)

INTRODUCTION

Chlorine (Cl₂) is a moderately soluble, highly reactive oxidant gas, used extensively for water purification, manufacturing of pharmaceuticals and chemicals and as a potent disinfectant. More than 25 million tons of chlorine is manufactured annually in the United States alone and the majority of this gas is transported by rail. Large quantities of chlorine gas have been released into the atmosphere following industrial accidents in production facilities or during transportation of chlorine to end users. Our data so far shows that exposure of rats to Cl₂ to either 187 or 400 ppm for 30 min severely decreases ascorbate levels in BAL and lung tissues and GSH/GSSG ratios in lung tissues. Furthermore, prophylactic intramuscular and intravenous injection of ascorbate, NAC and desferal increases ascorbate levels in BAL, normalizes GSH/GSSG values in lung tissue and decreases the extent of Cl₂ induced injury to the alveolar epithelium (in review).

In this series of experiments, we developed an aerosol system to deliver aerosolized ascorbate and deferoxamine into the lungs of mice, post Cl₂ exposure. Our results indicate that we can replete depleted ascorbate levels without damaging the alveolar epithelium.

PURPOSE

• Expose rats to Cl₂ (300 ppm for 30 min) in an environmental exposure chamber, and return to room air.
• One hour after exposure, deliver aerosolized ascorbate and deferoxamine via aerosol for one hour. Control animals received vehicle.
• Sacrifice the rats; perform BAL and remove lung tissues; obtain blood sample.
• Measure levels of ascorbate, reduced (GSH) and oxidized glutathione (GSSG) and urea concentrations in BAL, plasma and lung tissues.
• Calculate the volume of epithelial lining fluid and amounts of AA, GSH and GSSG in ELF and lung tissue.

RELEVANCE TO THE MISSION OF THE CounterACT PROGRAM

Chlorine gas was among the first chemical weapons to be used in modern warfare. During the last year Cl₂ cylinders were packaged with explosive materials by insurgents in at least four incidents in the Iraq war. In addition, nine people were killed and 250 injured in 2005 after a train crash in South Carolina in which 60 tons of liquefied chlorine was released to the atmosphere.

• Our previous findings suggest that prophylactic administration of ascorbate (Vitamin C) and deferoxamine (an iron chelator used to remove excess iron from the body) significantly reduce Cl₂ injury to the lung and restore arterial oxygenation to nearly normal levels.
• Herein, we compared the efficacies of various modes of administration of ascorbate and deferoxamine into the lungs of rats post Cl₂ exposure. The aim of these studies is to develop the most efficient and least invasive delivery mode of replenishing depleted antioxidant defenses in civilian and military personnel exposed to Cl₂.

Conclusions

Aerosol delivery of ascorbate in conscious rats post Cl₂ exposure restores depleted ascorbate levels in epithelial lining fluid and lung tissues without damaging the lungs.

Future Directions

We are currently evaluating whether restoration of depleted AA reverses Cl₂ injury to the alveolar epithelium. We also plan to use this system to deliver combinations of protective agents such as β2-agonists, nitrite and low molecular weight scavengers.