



STATEMENT

Synopsis of Research Report 110

HEALTH
EFFECTS
INSTITUTE

Effects of Metals Bound to Particulate Matter on Human Lung Epithelial Cells

INTRODUCTION

Inhaled particulate matter has been associated with both acute and chronic health effects. Concerns about these effects derive primarily from epidemiologic studies that associate short-term increases in particle concentration with increases in daily morbidity and mortality from respiratory and cardiovascular diseases. Over the past decade much research has been directed toward identifying plausible mechanisms linking particulate matter and pathophysiologic effects. Although progress has been made, many critical aspects are not understood. Thus, studies of the properties of particles that might induce pathologic effects are critical to establishing the mechanisms of particulate matter toxicity and to producing information necessary to target regulation of the sources that generate the most toxic particles.

Studies using laboratory animals have implicated metals associated with particulate matter in adverse health effects. Coal-fired power plants produce particulate residues called *fly ash*. Coal contains metals that vaporize during combustion and then condense on the surface of the ash. Inhaled coal fly ash could be a health hazard because metals solubilized from fly ash within lung cells may cause toxic reactions.

APPROACH

Dr Ann Aust and colleagues at Utah State University, the University of Utah, the University of California, Davis, and Ford Motor Company hypothesized that transition metals (metals that can participate in possibly toxic oxidative reactions) associated with particulate matter are released within lung epithelial cells and catalyze the formation of reactive oxygen species. Reactive oxygen species can stimulate epithelial cells to produce inflammatory mediators that contribute to lung inflammation and injury. The investigators focused their

study on coal fly ash that was produced in the laboratory and separated into four size fractions. (They also performed experiments using particles from gasoline and diesel exhaust, natural soils, and ambient Utah air.) This multifaceted study focused mainly on the ability of iron (the major transition metal in coal fly ash) to produce reactive oxygen species and inflammatory mediators in cultured lung epithelial cells.

RESULTS AND INTERPRETATION

This study was performed by experienced investigators with demonstrated excellence in the area of metal-catalyzed oxidative stress and particle-associated injury. The study was of high scientific quality, was well conceived and executed, and adds substantially to our knowledge of the biologic properties of particles.

Aust and colleagues found that more iron was released from the smaller particles than from larger ones. They confirmed that soluble extracts of coal fly ash generated reactive oxygen species *in vitro* and that transition metals were likely responsible. Further, the smallest particles, which were rich in iron, were the most active. The investigators then examined the effects of coal fly ash on human lung epithelial cells in culture. First, they demonstrated that coal fly ash particles entered the cells and stimulated synthesis of the protein ferritin. Ferritin binds iron and is produced in response to increasing iron levels; thus, its presence indicates that iron was released intracellularly and that iron was available to provoke an inflammatory response by forming reactive oxygen species. The investigators obtained indirect evidence for formation of intracellular reactive oxygen species by demonstrating that lung epithelial cells exposed to coal fly ash synthesized the inflammatory mediator interleukin-8. Ferritin and interleukin-8 production were stimulated to a greater degree by smaller

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particles than by larger ones. Thus, the investigators provided a plausible connection among the intracellular release of a transition metal from particles, formation of reactive oxygen species, and lung inflammation.

These findings may be important. To confirm their in vitro results, Aust and colleagues will measure ferritin levels in lung tissue and fluids from rats exposed to coal fly ash. The current results that smaller particles had greater effects supports the epidemiologic studies on the adverse effects of fine and ultrafine particles.

Other components or properties of particles have also been proposed to cause lung injury; therefore, there may be multiple mechanisms by which inhaled particles produce adverse health effects. Further research to identify particle characteristics (and sources) responsible for particulate matter toxicity is important for developing increasingly effective and appropriate air quality regulations, as noted in HEI Perspectives, *Understanding the Health Effects of Components of the Particulate Matter Mix: Progress and Next Steps*.

Particle Characteristics Responsible for Effects on Human Lung Epithelial Cells

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INVESTIGATORS' REPORT

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- Coal Fly Ash
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- Iron Species and Reactivity
- Significance

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Discussion and Conclusions

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Appendix B. Results of Additional Statistical Analyses

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HEALTH REVIEW COMMITTEE'S COMMENTARY

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- In Vivo and In Vitro Effects of Transition Metals Associated with Fly Ashes
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Technical Evaluation

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- Study Design and Methods

Results and Interpretation

- Physical and Chemical Properties of the Particles
- Biological Effects of the Particles

Discussion

Summary and Conclusions

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