**Controlled Human Exposure to PM and Gaseous Co-Pollutants**

Metabolomic Analysis: NIH Eastern Regional Comprehensive Metabolomics Resource Core (RTI RCMRC)

PI, RTI RCMRC Pilot Study: Susan Sumner, PhD., RTI International

PI, EPA: Robert Devlin, PhD., EPA

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**Abstract**

During the past decade, several epidemiological studies have reported statistically significant positive correlations between daily concentrations of ambient air particles and acutely increased mortality and morbidity. It has been estimated that 50,000 - 60,000 excess deaths in the U.S. each year may be attributable to ambient particles. Several panel studies have reported associations between fine PM and decreased heart rate variability and increased vascular markers of inflammation. In addition, recent controlled human exposure studies have reported that fine particles can increase pulmonary inflammation, decrease heart rate variability, and increase vascular factors of inflammation and blood coagulation. However, these latter studies only assessed the effects of particulate matter (PM). In the real world, people are simultaneously exposed to both gaseous pollutants (e.g. ozone, nitrogen dioxide) and particles. Recognition of this leads the National Research Council to list studies of PM and gaseous co-pollutants as one of the ten highest priorities in PM research.

One of these co-pollutants that frequently occur together with PM is nitrogen oxides (NOx), which is produced during combustion processes. NOx consists of nitric oxide (NO) and nitrogen dioxide (NO2). NO dominates near roadsides and peaks in morning rush hours while NO2 levels show less temporal and spatial variability. NO and NO2 concentrations may reach values over 1 ppm and 0.5 ppm respectively during smog situations.

NO2 is an oxidant capable of oxidizing and nitrating lipids and proteins and can cause cytotoxic effects on the cell membranes of epithelial cells as well as macrophages. Controlled exposure of healthy humans to 2 ppm NO2 reduced phagocytic capacity in macrophages. At similar concentrations controlled NO2 exposure produced small changes in large airway function and increased airway reactivity to methacholine. The inflammatory effects of NO2 may thus enhance the adverse effects of PM.

In this study we hypothesize that NO2 and PM2.5 affect the cardiopulmonary system beyond what either pollutant is capable of inducing by itself. Cardiopulmonary impairment will be assessed by measuring changes in bronchoalveolar lavage (BAL) neutrophils and cytokines, heart rate variability, and plasma factors involved in inflammation and coagulation.

**Goals**

This study is designed to provide the environmental aspects to support both the acquisition of study samples and the advancement of environmental chemical speciation information and data analysis needed. The aims of the study are as follows: 1) Do the metabolomics profiles appear to be impacted by exposure to PM and NO2+PM. 2) Are the metabolomics profiles related to the PM and NO2+PM distinct and 3) Which features (chemical or physical) of the PM and NO2+PM have the most significant impact on the metabolomic profiles?

The data required for the metabolomics analysis can be found in the accompanying files:

Procedures: 1. Controlled Human Exposure to PM and Gaseous CoPollutants Procedures.docx

1.a. GCMS Procedures Flowchart.pdf

1.b. GCMS Preparation of fatty acid methyl esters mixture.pdf

Study Design Table: 2. Controlled Human Exposure to PM and Gaseous CoPollutants Study Design.xlsx

Metadata: 3. Controlled Human Exposure to PM and Gaseous CoPollutants METADATA.xlsm

Raw Data: 4. Controlled Human Exposure to PM and Gaseous CoPollutants Raw GCMS Data.zip

Processed Data: 5. Controlled Human Exposure to PM and Gaseous CoPollutants Proc.xlsx

**Notes:**

Full sample preparation and instrumentation parameters are detailed in accompanying file **1. Controlled Human Exposure to PM and Gaseous CoPollutants Procedures.docx.** A flowchart detailing the sample preparation steps is located in accompanying file **1.a. GCMS Procedures Flowchart.pdf.** The preparation of the fatty acid methyl esters (FAME) mixture is located in accompanying file **1.b. GCMS Preparation of fatty acid methyl esters mixture.pdf**.

Factors listed in the study design are defined in the Variable Dictionary located in the accompanying file entitled **2. Controlled Human Exposure to PM and Gaseous CoPollutants Study Design.xlsx.** Available in the same file is information linking the Data File names to the Sample IDs.

Data files for each sample are generated by Leco’s ChromaTOF software and are exported in netCDF format . These files arelocated in the accompanying file entitled **4. Controlled Human Exposure to PM and Gaseous CoPollutants Raw GCMS Data.zip.**

The spreadsheet in accompanying file **5. Controlled Human Exposure to PM and Gaseous CoPollutants Proc.xlsx** has one data tab entitled BinBase Processed Data. The BinBase Processed Data shows raw output from BinBase. The height values have not been normalized.

**Reference**

O Fiehn, G. Wohlgemuth, M Scholz, T Kind, DY Lee, Y Lu, S Moon and B Nikolau: Quality control for plant metabolomics: reporting MSI-compliant studies. The Plant Journal2008; 53:691-704.