NYU Langone Health

Official Title:	Cardiometabolic Biomarker Improvements Associated With Reduction in Air Pollution Exposure Via Home Air Filtration
NCT Number:	NCT05379400
Study Number:	22-00045
Document Type:	Study Protocol and Statistical Analysis Plan
Date of the Document:	• September 14, 2023

NYU School of Medicine
NYU LANGONE MEDICAL CENTER

CARDIOMETABOLIC BIOMARKER IMPROVEMENTS ASSOCIATED WITH REDUCTION IN AIR POLLUTION EXPOSURE VIA HOME AIR FILTRATION.

Principal Investigators:	*Jonathan D. Newman, MD, MPH, FAHA, FACC* *Eugene Braunwald MD Assistant Professor of Medicine* *The Leon H. Charney Division of Cardiology* *And The Center for the Prevention of Cardiovascular Disease* *NYU School of Medicine*
Additional Investigators:	*Sharine Wittkopp, MD, PhD* *Post-Doctoral Fellow* *The Leon H. Charney Division of Cardiology* *NYU School of Medicine* *Terry Gordon, PhD* *Professor* *Department of Environmental Medicine* *NYU School of Medicine* *Jiyuan Hu, PhD* *Assistant Professor, Division of Biostatistics* *Department of Population Health* *Assistant Director, NYU Biostatistics Resource* *NYU School of Medicine* *Lorna Thorpe, PhD* *Professor* *Director of Division of Epidemiology* *Vice Chair of Strategy and Planning, Department of Population Health* *NYU School of Medicine*
NYULMC Study Number:	s22-00045
Funding Sponsor:	*NYU Clinical and Translational Science Institute*

Initial version: **01/25/2022**
Updated: 04/07/2022
Updated: 09/07/2023

Statement of Compliance

This study will be conducted in accordance with the Code of Federal Regulations on the Protection of Human Subjects (45 CFR Part 46), any other applicable US government research regulations, and institutional research policies and procedures. The Principal Investigator will assure that no deviation from, or changes to the protocol will take place without prior agreement from the sponsor and documented approval from the Institutional Review Board (IRB), except where necessary to eliminate an immediate hazard(s) to the study participants. All personnel involved in the conduct of this study have completed Human Subjects Protection Training.

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List of Abbreviations

BMI	Body Mass Index
BNP	Brain Natriuretic Peptide
BP	Blood Pressure
CBRD	Center for Biospecimen Research and Development
CDC	Center for Disease Control
CRP	C-reactive protein
CTSI	Clinical Translational Science Institute
EDTA	Ethylene diamine tetraacetic acid
EHR	Electronic Health Record
EPA	Environmental Protection Agency
GCP	Good clinical practices
GLP	Good Laboratory Practices
GMP	Good Manufacturing Practices
HEPA	High Efficiency Particulate Air
HIPAA	Health Insurance Portability and Accountability Act of 1996
ICAM-1	Intercellular Adhesion Molecule-1
ICMJE	International Committee of Medical Journal Editors
IL-6	Interleukin-6
IRB	Institutional Review Board
MCIT	Medical Center Information Technology
NCATS	National Center for Advancing Translational Science
NIH	National Institutes of Health
NYU	New York University
PAC	Portable Air Cleaner
PCSK9	Proprotein convertase subtilisin/kexin type 9
PHI	Protected Health Information
PI	Principle Investigator
PM	Particulate Matter
$PM_{2.5}$	Particulate Matter <2.5 μm in diameter
TNFα	Tumor Necrosis Factor-alpha
TP	Treating physician
TRB	Translational Research Building
VCAM-1	Vascular Cell Adhesion Molecule

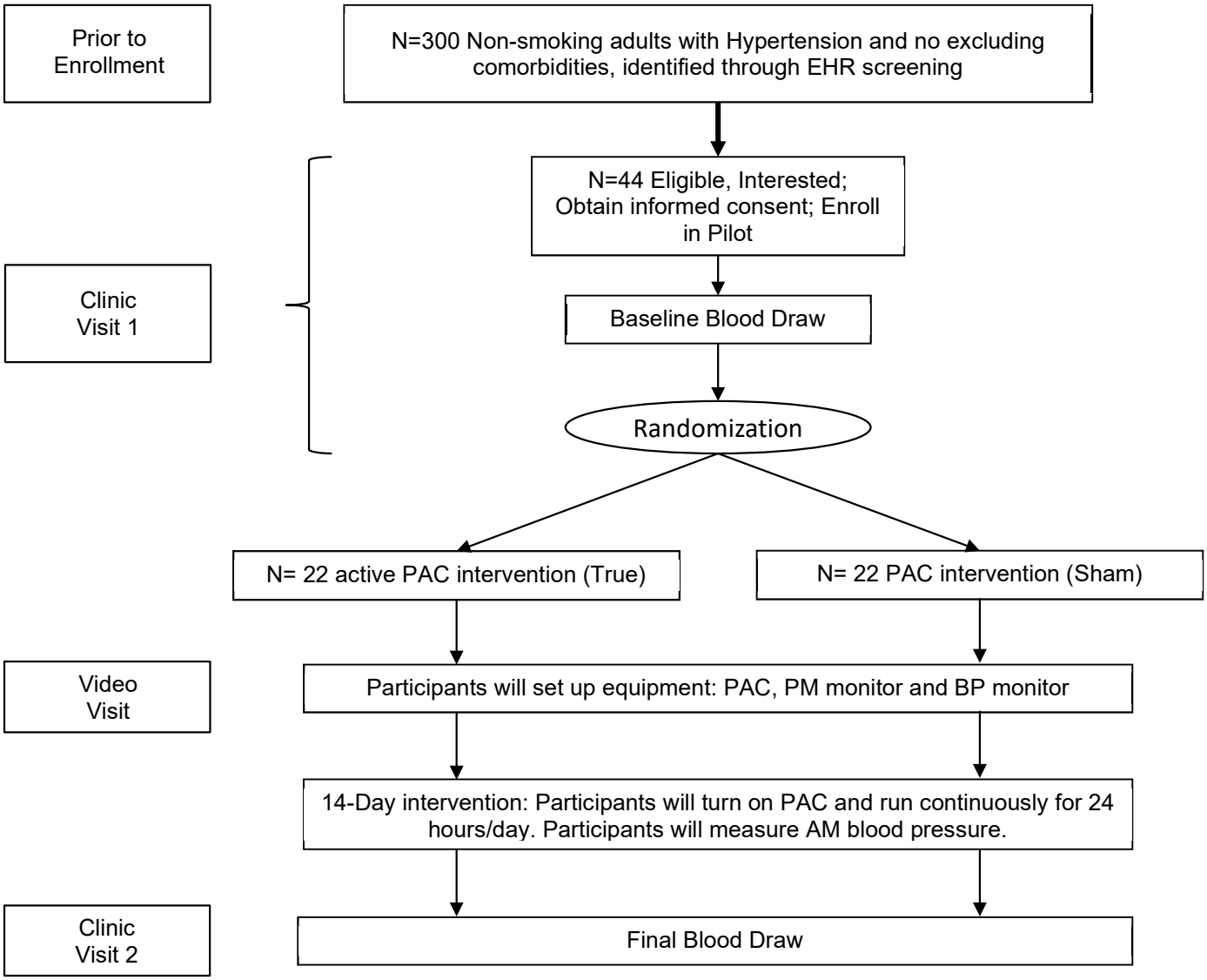
Protocol Summary

Title	*Cardiometabolic Biomarker improvements associated with reduction in air pollution exposure via home air filtration: a pilot trial.*
Short Title	*Cleaner air for better cardiac biomarkers*
Brief Summary	In this research, we will conduct a double-blind, sham-controlled, randomized trial to examine the impact on cardiometabolic biomarkers of air pollution exposure reduction using home portable air cleaners (PACs) in a cohort of current adult patients at NYU Langone Health. This study is funded by CTSI (NIH/NCATS UL1TR001445) with two primary aims and two secondary/exploratory aims. In **Aim 1**, we will evaluate associations with PAC use with *a priori* specified biomarker of inflammation, tumor necrosis factor alpha (TNFα). We will determine if 4 weeks of home air filtration with PACs compared to sham is associated with significant reductions in TNFα concentrations when measured before and after intervention. Blood samples will be collected from 44 study participants before and after 4 weeks of PAC use (true and sham) for biomarker concentration measurement. In **Aim 2**, we will determine if reductions in fine particulate matter (<2.5 μm in diameter, $PM_{2.5}$) as a continuous variable are associated with significant reductions TNFα concentrations over a four-week period. In **Exploratory Aim 1**, we will explore associations between PAC use and concentrations of cardiometabolic biomarkers using a targeted proteomic panel, the Olink Explore 384 Cardiometabolic panel, measured before and after 4 weeks of intervention. This will be a hypothesis-generating analysis to identify possible novel biomarkers and pathways that may have importance in the physiological responses and clinical outcomes associated with air pollution exposure. In **Exploratory Aim 2**, we will explore if biomarkers identified in Exploratory Aim 1 with significant reductions after 4 weeks of PAC partially mediate the association between reductions in $PM_{2.5}$ and changes in daily home blood pressure (BP) measurements. Participants will be given home BP monitors to transmit de-identified daily morning BP measurements to a secure database during the 4 week intervention period. Changes in morning systolic (BP) will be analyzed as the outcome of interest, with changes in biomarker concentrations evaluated as a mediator of the relations between pollution reduction and changes in BP.

Objectives	The primary objective of this research is to examine how PACs that reduce air pollution exposure affect cardiometabolic biomarkers and how those may mediate relationships with cardiovascular risk factors such as hypertension. Specific Aim 1: To determine if 4 weeks of home air filtration with PACs compared to sham is associated with significant reductions in TNFα concentrations when measured before and after intervention. Specific Aim 2: To determine if reductions in $PM_{2.5}$ are associated with significant reductions TNFα concentrations over a four-week period. Exploratory Aim 1: To identify novel biomarkers of the cardiometabolic effects of $PM_{2.5}$ exposure with PAC use with a targeted proteomic panel, the Olink Explore 384 Cardiometabolic panel, measured before and after 4 weeks of intervention. Exploratory Aim 2: To determine if novel biomarkers identified in Exploratory Aim 1 are mediators of the relationship between PAC use and home BP reductions after 4 weeks of PAC use.
Methodology	Randomized, double-blind, sham-controlled pilot study
Endpoint	Aim 1 primary endpoint: Change in circulating concentration of TNFα Aim 2 primary endpoint: Change in indoor $PM_{2.5}$ concentration Exploratory Aim 1 primary endpoint: Change in concentration of 384 circulating biomarkers associated with cardiometabolic risk Exploratory Aim 2 primary endpoint: Change in average AM systolic blood pressure over study period
Study Duration	4 weeks
Participant Duration	Maximum: 4 weeks
Population	We will perform a randomized, double-blind, sham-controlled trial of continuous (24 hours/day) bedroom PAC use in an outpatient cohort English and Spanish speaking adults ≥18 years of age with stable hypertension (defined as no medication changes in prior 30 days, systolic BP <160 mm Hg and either ≥ 130 mm Hg without antihypertensive medications, or diagnosis of hypertension in medical records). Per guidelines, this range is considered safe to monitor without changes in antihypertensive medications. Additional inclusion criteria require participants to measure home blood pressure twice daily, participate in video conference for home equipment setup, and ability to visit clinic for blood draws before and after the study period. Participants will be excluded if they are current smokers, currently have known coronary artery disease, have known systemic inflammatory conditions (such as rheumatoid arthritis, inflammatory bowel disease, cancer), or are currently taking any anti-inflammatory medications or medications that target inflammatory cytokines. We aim to enroll 44 participants, randomized in a 1:1 ratio to PAC with HEPA filtration versus sham filtration.

Study Sites	Participants will be consented and enrolled at NYU outpatient clinic facilities in Manhattan. At this visit, patients will have baseline blood drawn and will receive all study equipment. Participants will set up air pollution monitor and PAC in their homes with video chat guidance. PAC will be run continuously in participant bedrooms for 4 weeks. Participants will have follow-up visit to NYU Langone hospital clinical lab for post-intervention blood collection and equipment return.
Number of participants	To achieve 44 study participants, we will need to identify ≈300 eligible participants from the electronic health record (EHR) in NYU clinics. There will be 22 participants per arm (sham versus active filtration).
Statistical Analysis	For all Aims: descriptive summary statistics, such as mean and standard deviation (or median and inter quartile range) will be reported for continuous variables and counts and proportions for categorical variables. Graphical displays of all variables will be examined, with attention to assessing balance in these characteristics by intervention group, and with assessment of the distribution of variables, relevant to the choice of statistical tests. Statistical comparisons between the PAC and sham groups will be performed using a two-group two-sided t-test or nonparametric Wilcoxon test for continuous variables and Chi-square test for categorical variables. Distributions of biomarker concentrations will be examined and non-normally distributed concentrations will be log-transformed, as needed. The analysis of our primary outcome for **Aim 1** will use linear mixed-effects models with the change in prespecified biomarker, from pre-intervention to post-intervention, concentrations as the dependent variable and treatment arm as the independent variable. While randomization should obviate the need for additional adjustment, models will be adjusted for age, gender and BMI, as these are known to be related to inflammatory biomarker concentrations. These models use subject-specific random intercepts to account for heterogeneity between subjects and will account for within subject correlation between biomarker observations. For **Aim 2**, we will also utilize similar models incorporating $PM_{2.5}$ as the independent variable. This will be assessed as the difference between pre-intervention baseline $PM_{2.5}$ concentration and the time-weighted average concentration of $PM_{2.5}$ from the 14-day intervention period. For **Exploratory Aim 1**, we will evaluate the Olink panel results using mixed-effects models for the remaining biomarkers that were not prespecified. These will be the same models as for Aims 1 and 2, adjusted for age, gender, and BMI. For **Exploratory Aim 2**, we will evaluate associations of PAC use and $PM_{2.5}$ reduction with measured AM systolic blood pressure in this clinical cohort. We will perform mediation analysis using biomarkers with significant associations in Exploratory Aim 1 as mediators of the relations between $PM_{2.5}$ reduction and change in blood pressure.

Schematic of Study Design



1 Key Roles

Principal Investigator
Jonathan Newman, MD, MPH, Eugene Braunwald M.D. Assistant Professor of Medicine (Cardiology)
Division of Cardiology, Department of Medicine
Center for the Prevention of Cardiovascular Disease
NYU School of Medicine
TRB Ste 853, NYU School of Medicine
227 E 30th, NY, NY 10016
212.263-9393
Jonathan.Newman@nyulangone.org

co-Investigator
Sharine Wittkopp, MD, PhD
Division of Cardiology, Department of Medicine
NYU School of Medicine
TRB Ste 817A, NYU School of Medicine
227 E 30th, NY, NY 10016
646-501-8212
Sharine.Wittkopp@nyulangone.org

co-Investigator
Terry Gordon, PhD, Professor
Department of Environmental Medicine, NYU School of Medicine
341 East 25th St, NY, NY 10010
646-754-9450
Terry.Gordon@nyulangone.org

co-Investigator
Jiyuan Hu, PhD
Assistant Professor, Division of Biostatistics
Department of Population Health
Assistant Director, NYU Biostatistics Resource
180 Madison Avenue, 2nd Flr, NY, NY 10016
6465013639
Jiyuan.Hu@nyulangone.org

co-Investigator
Lorna Thorpe, PhD MPH, Professor
Director of Division of Epidemiology, Vice Chair of Strategy and Planning, Department of Population Health, NYU School of Medicine
650 First Avenue, room 516, New York, NY 10016
212-263-3327
Lorna.Thorpe@nyulangone.org

2 Introduction, Background Information and Scientific Rationale

2.1 Background Information and Relevant Literature

Fine particulate matter air pollution (PM≤2.5 μm in diameter, $PM_{2.5}$) exposure is associated with increased blood pressure.[1, 2] Proposed mechanisms include increases in inflammation and oxidative stress leading to increased sympathetic tone and impaired vasomotor regulation.[3, 4] We have identified gene expression changes in inflammatory pathways in association with increased particulate matter

pollution.[5] Additionally, downstream changes were shown in protein expression of TNFα and other inflammatory biomarkers.[6-8] Controlled human exposure studies show that exposure to air pollution increases TNFα;[9] *in vivo* and *in vitro* studies demonstrate that TNFα promotes the vascular inflammation and remodeling linked with increases in blood pressure and the development of hypertension.[10] Given this spectrum of evidence, these studies support a probable role for air pollution in increasing TNFα and the hypothesis that increases in TNFα may be related to increases in blood pressure with fine particulate matter air pollution exposure.

While it is clear that increases in $PM_{2.5}$ exposure are associated with increases in inflammation, ***evidence is less well-established that reducing $PM_{2.5}$ exposure will reduce inflammation.*** Natural experiments in Beijing during the 2008 Olympics showed that reductions in $PM_{2.5}$ due to regulations for the event were associated with reductions in concentrations of circulating inflammatory biomarkers compared to the months immediately before and after.[11] There are also limited studies of reducing cardiovascular risk factors by decreasing particulate matter exposure with the use of HEPA filtration by portable air cleaners (PACs). For example, in a meta-analysis of the effect of PAC use on blood pressure, our group showed a significant reduction in systolic blood pressure by ≈4 mm Hg over a median of 13.5 days of intervention versus sham.[12] More recently, we also showed by meta-analysis that residential PAC use by adults is associated with reductions in circulating concentrations of CRP (10 studies, total N=570) and IL-6 (9 studies, total N=379).[13] In these studies, active PAC use was associated with statistically significant 7% lower CRP and 13% lower IL-6 vs. sham control. TNFα was lower by 2%; however, this biomarker had insufficient data to reach statistical significance (5 studies, N=215). Multiple biomarkers have data supporting their associations with air pollution exposure. For example, recent studies have shown that $PM_{2.5}$ exposure was associated with significant changes in Angiopoetin-1, ICAM-1 and VCAM-1,[14] and increased expression of CRP, IL-6, TNFα, endothelin-1, fibrinogen, p-selectin.[6-8] PCSK9 is an important pharmacologic target that has been shown to increase in association with increased particulate matter exposure.[15] Soluble platelet selectin (sP-selectin) has been shown to increase with PM exposure; a small trial of 29 adults showed that sP-selectin decreased with PAC use versus sham.[16] BNP increases were shown in human exposure trials using diesel exhaust, with reductions in BNP when exhaust was filtered.[17] The data from these studies support connections with exposure and biomarker outcomes, but there are limited data from few small studies describing associations with PAC use. Given the important role in cardiovascular disease, and data showing relationships with pollutant exposure, we will primarily evaluate changes in TNFα in association with PAC use versus sham; and also will explore associations with Angiopoetin-1, ICAM-1, VCAM-1, PCSK9, sP-selectin, and BNP and other panel biomarkers. ***There is a dearth of data on how personal interventions to reduce air pollution exposure affect cardiometabolic biomarkers and how those may mediate relationships with cardiovascular risk factors such as hypertension.*** To address this gap we have proposed a randomized, double-blind, sham-controlled pilot trial to evaluate the changes in inflammation over 4 weeks of PAC use.

2.2 Rationale

Because PAC use has been associated with decreases in concentrations of circulating CRP and IL-6, we hypothesize the following: (1) Among adults with hypertension recruited from an outpatient clinical setting, 4 weeks of continuous home PAC use, compared with sham PACs, will be associated with decreased concentrations of TNFα when measured before and after intervention; (2) Reductions in measured $PM_{2.5}$ from PAC use will be associated with decreased concentrations of circulating TNFα.

2.3 Potential Risks & Benefits

2.3.1 Known Potential Risks

This is a minimal risk study, with no physical risks associated with the study design. Potential minimal risks are as follows:

(1) **Risk of PAC use:** In our published review of the effect of personal air cleaners (PACs) on blood pressure control, no risks were reported.[12] While it is possible that use of a PAC might disrupt sleep, it is unlikely. In fact, frequently PACs have been used to generate “white noise” associated with improved sleep latency and duration in a variety of settings.

(2) **Risk of monitoring air pollution:** There are no known risks to participants with monitoring the amount of air pollution exposure. At conclusion of the 3-month study, data on air pollution exposure will be provided to study participants. There is no risk to participants in ongoing air pollution exposure during this time interval.
(3) **Risk of blood collection.** There are minimal risks associated with a standardized venipuncture for blood collection. These risks include pain, discomfort and superficial bleeding or bruising. All participants in the clinical cohort will provide consent for standard blood collection, in which these minimal risks will be described. For participants in the clinical cohort, up to <15 ml (≈1 tablespoons and ≈3% of a standard blood donation) of blood will be collected at the baseline visit and 4-week visit. The total amount of blood collected will be <30 mL for the main study. For participants opting into biospecimen storage, an additional ≈10mL will be drawn at each visit for a total of <50 mL for the entire study. This presents minimal risk to study participants.
(4) **Risk of monitoring home blood pressure and ambulatory blood pressure.** There are no known direct risks to participants associated with the use of home blood pressure monitors are ambulatory monitors. These devices are routinely prescribed for self-monitoring and clinician-assisted decision making for hypertension. There is a potential risk of increased anxiety due to self-monitoring of blood pressure. Study coordinators will discuss any concerns regarding home blood pressure monitoring at video visit with study participants and will refer any concerns on recorded blood pressure values to participants' managing physician.
(5) **Risk of discomfort** with staff visualizing participants' homes during video visit for equipment setup
(6) **Loss of time** related to study participation: to minimize this risk, staff will schedule video visits at participants' preferred times, and coordinate 4-week intervention period to facilitate clinic visits at convenient times for participants.

2.3.2 Known Potential Benefits

This research will significantly advance our understanding of the relationship between PAC use and inflammation, and potentially with blood pressure health outcomes. Using a sham-controlled design, the proposed study will provide robust evidence to enhance the existing knowledge on this subject and inform future research questions.

Though individual study participants may experience the above mentioned direct potential risks through their participation in this study, study participants may also benefit from the study: study participants will receive and keep their own personal PAC and BP monitor, these may result in lasting improvements in participant health via continued air filtration or enhanced awareness of their BP. Participants will also receive a small remuneration for each blood draw.

3 Objectives and Purpose

3.1 Primary Objective

The primary objective of this research is to examine the effectiveness of PAC use in improving inflammation related to cardiovascular disease.

Specific Aim 1: To determine if 4 weeks of home air filtration with PACs compared to sham is associated with significant reductions in TNFα concentrations when measured before and after intervention.

Specific Aim 2: To determine if reductions in $PM_{2.5}$ as a continuous variable are associated with significant reductions TNFα concentrations over a four-week period.

Exploratory Aim 1: To explore associations between PAC use and concentrations of cardiometabolic biomarkers using a targeted proteomic panel, the Olink Explore 384 Cardiometabolic panel, measured before and after 4 weeks of intervention.

Exploratory Aim 2: To explore if changes changes in TNFα and other novel cardiometabolic biomarkers partially mediate the effects of PACs on home blood pressure.

4 Study Design and Endpoints

4.1 Description of Study Design

In this research, we will conduct a randomized, double-blind, sham-controlled pilot trial to study the effect of PAC use on the concentrations of circulating biomarkers of inflammation in outpatient adults within the NYU Langone Health system.

Aim 1: To determine if 4 weeks of home air filtration with PACs compared to sham is associated with significant reductions in TNFα concentrations when measured before and after intervention. Data collection will begin Q2 2022 with a total of two time points 4 weeks apart, pre- and post-intervention. The study design utilizes purposeful selection of 74 adults with stable hypertension within the NYU Langone heath system outpatient cohort. We will define stable hypertension as no medication changes in prior 30 days, systolic BP <160 mm Hg and either ≥ 130 mm Hg without antihypertensive medications, or diagnosis of hypertension in medical records. Per guidelines, this range is considered safe to monitor without changes in antihypertensive medications. At the initial clinic visit, blood will be collected and stored for analysis. We will measure circulating concentrations of TNFα using the Olink Explore 384 Cardiometabolic panel for this *a priori* specified analysis; this panel will also measure the concentrations of other biomarkers that we will explore further in Exploratory Aims 1 and 2. The Olink analysis uses a Proximity Extension Assay that has been shown to have high specificity and accuracy to allow for assessment of changes in concentrations between repeated samples over time.[18, 19]

We will randomize participants to receive PAC with either true or sham filtration (HEPA filter removed). Equipment will be shipped to participant homes or given to participant at clinical visit along with written instructions for setup and use. Active and sham PACs, and pollution monitors, will be positioned in the bedroom where each participant sleeps. We will video conference with participants for initial setup of equipment, though prior data suggests that written instructions along are effective in aiding participants in this process. We will use a commercially available portable air cleaner with a “true HEPA” filter for the true arm. For the sham arm, the same model will be used with HEPA filter removed, and has identical appearance and sound. After enrollment and baseline blood draw, Home BP will be monitored using commercially available device with a paired data hub that transmits BP measurements to an online dashboard in real time. We will monitor participant adherence to PAC use with electricity consumption monitors. Participants will be scheduled for a clinic visit after 4 weeks of PAC use to return data hubs, electricity monitors and pollution monitors to the study team, and will undergo post-treatment blood draw. A second blood draw will be performed and blood will be stored for analysis. Participants will receive reimbursement after equipment is returned and second blood draw is complete. Participants randomized to sham PAC arm will receive HEPA filters to install and keep.

Aim 2: To determine if reductions in $PM_{2.5}$ as a continuous variable are associated with significant reductions TNFα concentrations over a four-week period. For Aim 2, the design is identical to above; however in addition, we will collect $PM_{2.5}$ data using commercially available PM sensors placed in the same bedroom as PAC. While it has been shown that PACs significantly lower indoor $PM_{2.5}$ concentrations, these data will allow us to examine the effect of PM as a continuous variable in addition to the binary variable of PAC use (active versus sham). Commercially available PM monitors will record indoor $PM_{2.5}$ concentrations using dual (backup) sensors, which store measurements on internal SD cards. Commercially available PM sensors have been well validated and use laser particle counters to count the number of particles at varying sizes with a sampling frequency of 2 minutes. PM monitors will be returned and data will be retrieved from SD cards at the post-intervention blood draw visit. These data will be measured for the entire cohort.

Exploratory Aim 1: To explore associations between PAC use and concentrations of cardiometabolic biomarkers using a targeted proteomic panel, the Olink Explore 384 Cardiometabolic panel, measured before and after 4 weeks of intervention. For this Aim, we will utilize additional data generated from the Olink panel. The Olink Explore Cardiometabolic panel is for research use only and will not be used toward FDA approval or diagnostic purposes. It will generate no clinically actionable results. The panel measures 369 protein biomarkers associated with cardiovascular and metabolic disease. This panel has been used to investigate cardiovascular disease treatment and prevention in various adult populations. [18, 19] [18, 19] [18,19] [18, 19] The pathways represented in the cardiometabolic panel include angiogenesis, cell activation, cell adhesion, coagulation, complement activation, extracellular matrix organization, response to hypoxia, and others. We will explore associations between PAC use (active vs. sham) and changes in circulating concentrations of Angiopoetin-1, ICAM-1, VCAM-1, PCSK9, sP-selectin, and BNP and other panel biomarkers. This Exploratory Aim will require no additional action on the part of participants.

Exploratory Aim 2: To explore if changes in TNFα and other novel cardiometabolic biomarkers partially mediate the effects of PACs on home blood pressure. For this Aim, participants will monitor blood pressure daily during the 4-week intervention period using commercially available BP monitor with a paired data hub that transmits BP measurements to an online dashboard in real time. Participants will receive written and verbal instructions for proper BP recording technique at the baseline visit. They will record two measurements in a seated position each morning and evening during the study periods. The outcome of interest will be the mean of two AM systolic blood pressure measures. We will perform mediation analysis to analyze the direct and indirect effects of changes in significant biomarkers and $PM_{2.5}$ changes on AM systolic BP. Based on our own pilot study of the effect of PAC use on AM systolic BP, we anticipate up to 30% missing BP measurements. We surveyed our prior pilot cohort regarding reasons for missing measurements and the predominant reported reason was forgetting; participants noted that they would have liked daily reminders. We will offer participants the option to receive daily text message reminders to record BP. These have been reported to be effective in other studies of home BP as well.

4.2 Study Endpoints

4.2.1 Primary Study Endpoints

Aim 1 and Aim 2: The primary study endpoint will be % change in concentration of TNFα over the 4-week intervention period calculated as:

$$\frac{([post-intervention]-[pre-intervention])}{[pre-intervention]} * 100\%$$

Because the baseline biomarker concentration for an individual is compared to post-intervention irrespective of range, percent change is a useful metric to compare reported results across assays.

4.2.2 Exploratory Endpoints

Exploratory Aim 1: The endpoints for Exploratory Aim 1 will be the % change in concentration of biomarkers from the Olink Explore Cardiometabolic panel over the 4-week intervention period calculated as:

$$\frac{([post-intervention]-[pre-intervention])}{[pre-intervention]} * 100\%$$

Exploratory Aim 2: The endpoint for Exploratory Aim 2 will be the change in AM systolic BP (mm Hg) as a function of the change in $PM_{2.5}$ (in µg/m3) from baseline.

5 Study Enrollment and Withdrawal

5.1 Inclusion Criteria

To be eligible to participate in the pilot, individuals must meet all of the following criteria:

1. Participant is ≥18 years old
1. Participant and members of household are reported to be non-smokers (defined as no household members are currently known to smoke cigarettes or use other tobacco products including e-cigarettes)
2. Participant is to understand/speak English or Spanish
3. Participant can understand study procedures and give informed consent
4. Participant has stable hypertension: no medication changes in prior 30 days, systolic BP <160 mm Hg and either ≥ 130 mm Hg without antihypertensive medications, or diagnosis of hypertension in medical records
5. Able to measure home blood pressure twice daily
6. Able to participate in video conference for home equipment setup
7. Able to visit clinic for blood draws before and after the study period
8. Do not intend to sleep anywhere outside of primary bedroom for more than 48 consecutive hours or a total of 7 days during the study period.
9. Ability to lift, or access to assistance with lifting, 20 lb to set up PAC in bedroom.

5.2 Exclusion Criteria

Any individual who meets any of the following criteria will be excluded from participation in the pilot:

1. Participants who are unable to provide a minimum of at least one blood pressure measurement per day
2. Participants with average home blood pressure monitor readings of a systolic blood pressure (SBP) >160 mmHg over any 10-day period during the study. This will be considered evidence of uncontrolled hypertension and will require study termination.
3. Participants with known coronary artery disease
4. Participants with known systemic inflammatory conditions (such as rheumatoid arthritis, inflammatory bowel disease, cancer)
5. Participants currently taking any anti-inflammatory medications or medications that target inflammatory cytokines
6. Pregnancy
7. Participants with known or suspected Covid-19 in the prior 30 days
8. Participants with post-covid sequelae (a.k.a “Long covid”)
9. Participants who are already utilizing HEPA filtration (PAC or within HVAC) in bedrooms

5.3 Vulnerable Subjects

The study does not intend to enroll children, pregnant women, prisoners, or other vulnerable populations.

5.4 Strategies for Recruitment and Retention

To achieve 44 study participants who complete the pilot, we will need to identify ≈300 eligible participants from the electronic health record (EHR) in NYU clinics and plan to enroll and randomize 44 participants. Utilizing support from DataCore and NYU Medical Center Information Technology, we will use SlicerDicer and other Epic tools to review EHR for eligibility. The team will request MCIT DataCore to generate a report identifying potential subjects that meet criteria and have upcoming office visit appointments. The study team, including principal investigator, co-investigators, and research assistants, will have access to the Epic search results and verify eligibility of potential subjects. The search results will include PHI- such as medical record numbers. Patients will then be contacted by phone, email, or MyChart portal using an IRB approved message or script. Subjects will also be approached during office visits to introduce the research project and be invited to participate in the study. In addition, the Clinical and Translational Science Institute (CTSI) Recruitment and Retention Core at NYU will be engaged to provide expert consultation.

Interested and eligible participants that have an upcoming clinic appointment will be met at the visit for consent and enrollment. If no clinic visits are upcoming, participants will be invited for a research only

visit. Participants will meet with a research coordinator to review eligibility and provide informed consent. Informed consent will include specific language regarding laboratory testing for the Olink biomarker panel and a separate consent subsection for participation in providing samples for future research. Study discussion and consenting will occur following a clinic visit in a secure private location in either the outpatient clinics or Clinical and Translational Research Institute of NYU Medical Center. Participant will be randomized to true or sham arm of the trial and baseline blood draw will occur at this clinic visit. Blood will be collected and stored for analysis. Participants will receive partial reimbursement for the initial blood draw, and the remainder of remuneration will be giving at the final blood draw/equipment return.

Inclusion of Women and Minorities: Women will be included as study participants (approximately 64% of enrolled participants). Minorities will also be included as study participants. Among NYU Langone Health outpatients, African Americans, Hispanic/Latinx, and Asian outpatients represent 16%, 17%, and 5%, respectively, of the potential pool of study participants.

5.4.1 Use of DataCore/Epic Information for Recruitment Purposes

This study will utilize Epic to identify subjects. We will work with DataCore and NYU Medical Center Information Technology to identify ≈300 eligible participants from the EHR in NYU clinics. We will use SlicerDicer and other Epic tools to review EHR for eligibility. The team will request MCIT DataCore generate a report identifying potential subjects that meet criteria and have upcoming office visit appointments. The study team, including principal investigator, co-investigators, and research assistants, will have access to the EPIC search results and verify eligibility of potential subjects.

Any recruitment information sent by email will utilize Send Safe email.

Once potential subjects have been identified, the study team will notify the treating physician (TP) that they have patients eligible to participate as follow:

- TP has been notified that the study team will contact potential subjects directly, by letter, phone, email, or the MyChart portal etc.

Once contact is made, approved recruitment language will be used to communicate the reason they are being contacted and subjects will be asked if they are interested in participating in this specific study. Should the potential subjects agree, the study team will provide the subjects with information regarding the next steps for participation.

If a subject requests information regarding opting out of further recruitment for all research, subjects will be directed to contact research-contact-optout@nyumc.org or 1-855-777-7858.

5.5 Study Participation

For the initial clinical visit, participant's participation will last approximately 30 minutes for consent and blood draw. The video visit for equipment set up will require 15 minutes. Daily participation will consist of 5-7 minutes for BP monitoring each morning and evening. The final clinical visit for the post-intervention blood draw will last approximately 10-15 minutes.

5.6 Total Number of Participants and Sites

This pilot study will be conducted at NYU Langone Health Main Campus. Recruitment will end when we have consented and enrolled 74 adults, of which we expect 90% to complete the trial.

5.7 Participant Withdrawal or Termination

5.7.1 Reasons for Withdrawal or Termination

This is a minimal risk study, so we do not expect participants to withdraw for a safety related issue. However, participants are free to withdraw from participation in the study at any time upon request. If patients need to sleep elsewhere than their own bedroom for any unanticipated reason during the trial,

they should notify the research team and study endpoints (exposures and SBP) will be excluded from analyses during this period. Participants that unexpectedly require sleeping 2 or more consecutive nights away from their home (or a total of 7 days altogether) during the study period will need to drop from the study and their data will not be used in analyses. They can re-enroll at a later date.
Reasons for withdrawal may be moving from the area or simply not interested in participating further.

5.7.2 Handling of Participant Withdrawals or Termination

Participation is voluntary and all study participants may withdraw from the study at any time without penalization. Participants may request removal and destruction of their blood samples from storage for future research without withdrawal from the remainder of the study. Participants may request termination from the study separate from withdrawal from future research.

No replacement of participants who withdraw or discontinue early will occur. Data including date and reason for withdrawal will be recorded.

A participant is considered lost to follow-up after 10 attempts to contact them by phone and 10 attempts by Epic messaging.

5.7.3 Premature Termination or Suspension of Study

This study may be suspended or prematurely terminated if there is sufficient reasonable cause. Written notification, documenting the reason for study suspension or termination, will be provided by the suspending or terminating party to the study principal investigators (Newman, Wittkopp). If the study is suspended or prematurely terminated the PIs will promptly inform the IRB and will provide the reason(s) for the termination or suspension.
Circumstances that may warrant termination or suspension include, but are not limited to:

- Insufficient compliance to protocol requirements
- Loss of institutional political will to examine health effects of the national policy
- Determination that the study procedures are too invasive

Study may resume once concerns about safety, protocol compliance, data quality are addressed and satisfy the sponsor and/or IRB.

6 Study Agent (Study drug, device, biologic, vaccine etc.) and/or Procedural Intervention

This study will utilize a portable air cleaner as an intervention for the potential reduction of air pollution-associated inflammation. This commercially available device has no significant risks. We plan to use a commercially available PAC with a “true HEPA” filter for the true arm. For the sham arm, the same model will be used with HEPA filter removed, and has identical appearance and sound. We will use a PAC designed to filter air in rooms up to approximately 350 ft^2, with minimal noise on the lowest setting.

6.1 Study Agent(s) and Control Description

This study will utilize a sham PAC as a control. PAC will be delivered to participants as active or sham PAC in blinded fashion. The study team with receive the new devices and remove filters from sham PACs. Filters will be stored in our research office until the participant completes the protocol, at which time participants will return study equipment at receive filters to keep. Filter status indicator lights on the PAC will be covered with black electrical tape on all devices (active and sham) to preserve blinding.

6.1.1 Acquisition

PACs will be ordered online and delivered to the study team research office for preparation.

6.1.2 Formulation, Appearance, Packaging, and Labeling

This is a commercially available product and will be delivered to participants in original packaging, which will have been opened to prepare and blind devices.

6.1.3 Preparation

Commercially available PACs contain carbon filters that reduce volatile organic carbons and can reduce odors. Because of this, we will remove the carbon filters from all air purifiers to preserve blinding. Sham PACs will also have HEPA filters removed. This will be done by unblinded research staff and devices will be labelled with an ID number to preserve blinding of the rest of the research team.

6.1.4 Duration of Therapy

The PACs will be run continuously (24 hours per day) in participants' bedrooms for 4 weeks.

7 Study Procedures and Schedule

7.1 Study Procedures/Evaluations

7.1.1 Study Specific Procedures

History will be obtained via participant medical records and confirmed with questionnaire. Medical comorbidity information collected will include: history of hypertension, duration of hypertension, history of diabetes; history of inflammatory conditions including Crohn's disease, ulcerative colitis, rheumatoid arthritis, vasculitides such as Kawasaki disease and giant cell arteritis, cancer and current status (no evidence of disease, ongoing treatment, cured, etc). We will also collect current prescription medications and assess for any planned medication changes during the study period. We will collect blood pressure, BMI and waist circumference at the initial visit. We will collect <15 mL of blood for evaluation of inflammatory biomarkers at both the initial visit and the final visit. An additional ~10 mL of blood will be drawn from participants opting in to storage for future research.

7.1.2 Standard of Care Study Procedures

Regular standard of care procedures will be performed by clinical providers at the initial clinical visit and data will be recorded to RedCAP for research purposes by research staff at the time of enrollment. These data include measurement of blood pressure, height and weight and waist circumference. Medication reconciliation performed for the clinical visit will be confirmed by research staff as well.

7.2 Laboratory Procedures/Evaluations

7.2.1 Other Assays or Procedures

We will evaluate biomarkers of inflammation using the Olink Explore 384 Cardiometabolic panel. This panel will be performed by Olink. The Olink analysis uses a Proximity Extension Assay that has been shown to have high specificity and accuracy to allow for assessment of changes in concentrations between repeated samples over time.

7.2.2 Specimen Preparation, Handling, and Storage

Blood samples will be processed for both cardiometabolic biomarker measurement and storage for future research. Samples for Olink will be collected into EDTA coated tubes and centrifuged within 1 hour and samples will be placed in 96-well PCR plate (ThermoFisher Scientific #AB0800 with MicroAmp seal #4306311 from ThermoFisher Scientific). Samples will be randomized within plates using random-number generator and stored at -80C until shipped on dry ice to Olink for analysis of biomarker concentrations. Olink will not receive any participant identifying information. The remainder of blood will be stored in a secured laboratory in the Cardiology Division Science Building Laboratories.

7.2.3 Specimen Shipment

Specimens will be stored at -80C until shipped on dry ice to Olink when 96-well plates are filled until all data is collected. After all data is collected, remaining specimens for participants who have not opted in to future research will be discarded by the lab.

7.3 Study Schedule

7.3.1 Screening

Screening will occur with support from DataCore/MCIT

- Obtain list of potential participants in NYU Langone Health System
- Review list of potential participants to identify those with upcoming clinical visits (during Q2-3 of 2022)
- Review potential participant charts to confirm likely eligibility
- Message eligible participants' providers to coordinate staff visit at clinical appointment
- MyChart messages will be used to contact potential research participants.

7.4 Enrollment/Baseline

Enrollment/Baseline Visit (Visit 1)

- Verify inclusion/exclusion criteria
- Answer questions from potential participants regarding the air monitors, PACs and BP monitors to verify interest in participating
- Obtain written consent from eligible participant
- Schedule study visits (video visit for setup and clinical visit 2 for second blood draw) for individuals who are eligible and available to participate for the full duration of the study
- Record participant BP, height, weight, and waist circumference if not done as part of clinical visit
- Collect blood for baseline biomarker measurement pre-intervention: <15 mL for all, additional ~10 mL for participants opting into future research.

7.5 Video Visit

Video visit for equipment setup

- Conduct video visit at a mutually agreeable time to instruct participants in equipment setup
    - Place PAC in an acceptable location in bedroom with recommended air space surrounding PAC, and PAC plugged into electricity monitor
    - Place PM monitor in an acceptable location in bedroom with adequate space between PAC and PM monitor to ensure representative sample
    - Connect PM monitor to WiFi if available
- Supervise participant self-measuring BP once to ensure real-time data transmission
- PAC must be turned on ≤7 days from baseline blood draw (Visit 1)

7.6 Final Study Visit

Final Study Visit (Visit 2: 4 weeks after PAC turned on)

- Participants return study equipment: PM monitors, BP monitor data hubs, electricity monitors
- Collect blood for post-intervention biomarker measurement: <15 mL for all, additional ~10 mL for participants opting into future research.
- Final blood draw must occur ≤35 days from turning on PAC

7.7 Withdrawal Visit

Subjects may withdraw from the study at any time and visit will be scheduled to return any study equipment that has been received (PM monitors, BP monitor data hubs, electricity monitors). No withdrawal visit is required for withdrawal from future research.

7.8 Unscheduled Visit

Participants may request an unscheduled video visit to troubleshoot equipment-related issues if needed.

8 Assessment of Safety

This study involves the use of commercially available products associated with no known risks to consumers. The Principle Investigator and Co-Investigator, Drs. Newman and Wittkopp, will monitor data safety of the overall study. We will review safety events related to blood pressure monitoring and blood draws at weekly team meetings with biweekly participant reviews complied by study coordinators. Given the established safety of all commercially available equipment used in this protocol, there are no predefined stopping rules for the study (no superiority/futility). After data safety assessments, study team will be notified via email of any changes in protocol.

9 Clinical Monitoring

Clinical site monitoring is conducted to ensure that the rights and well-being of human subjects are protected, that the reported trial data are accurate, complete, and verifiable, and that the conduct of the trial follows the currently approved protocol/amendment(s), and with applicable regulatory requirement(s). This is a single-site study; data, training, procedures, and practices will be monitored continuously throughout the study period by study PIs. A quality management plan will be developed if needed.

10 Statistical Considerations

10.1 Statistical Hypotheses

Aim 1:
Hypothesis: Among adults with hypertension recruited from an outpatient clinical setting, 4 weeks of continuous home PAC use, compared with sham PACs, will be associated with decreased concentrations of TNFα when measured before and after intervention.

Aim 2:
Hypothesis: Reductions in measured $PM_{2.5}$ from PAC use will be associated with decreased concentrations of circulating TNFα

10.2 Analysis Datasets

These analyses will be conducted as Intention-to-Treat dataset. Per-Protocol Analysis will be explored if electricity monitor data indicates poor compliance with PAC usage (if electricity monitor recordings indicate kilowatt-hour usage that is lower than expected by 50%.

10.3 Statistical Methods

Aim 1 Analysis.

Descriptive summary statistics, such as mean and standard deviation (or median and inter quartile range) will be reported for continuous variables and counts and proportions for categorical variables. Graphical displays of all variables will be examined, with attention to assessing balance in these characteristics by intervention group, and with assessment of the distribution of variables, relevant to the choice of statistical tests. Statistical comparisons between the PAC and sham groups will be performed using a two-group two-sided t-test or nonparametric Wilcoxon test for continuous variables and Chi-square test for

categorical variables. Distributions of biomarker concentrations will be examined, and non-normally distributed concentrations will be log-transformed, as needed.

The analysis of our primary outcome will use linear mixed-effects models with the change in prespecified biomarker, from pre-intervention to post-intervention, concentrations as the dependent variable and treatment arm as the independent variable. While randomization should obviate the need for additional adjustment, models will be adjusted for age, gender and BMI, as these are known to be related to inflammatory biomarker concentrations.[20, 21] These models use subject-specific random intercepts to account for heterogeneity between subjects and will account for within subject correlation between biomarker observations. The proposed model is:

$$\Delta\, TNF\alpha = \beta_1 + \beta_2(PAC\ arm) + \beta_3(age_i) + \beta_4(gender_i) + \beta_4(BMI_i) + e_i$$

We will also utilize similar models incorporating $PM_{2.5}$ as the independent variable in place of PAC arm. This will be assessed as the difference between pre-intervention baseline $PM_{2.5}$ concentration and the time-weighted average concentration of $PM_{2.5}$ from the 14-day intervention period. For our exploratory aim we will evaluate the Olink panel results using mixed-effects models for the remaining biomarkers that were not prespecified. Additionally, we will explore associations of PAC use with measured blood pressure in this clinical cohort that likely differs from our current pilot cohort recruited from a community setting. Because this pilot will integrate with our larger R01 trial, for which blood pressure is the primary outcome of interest, we will incorporate this biomarker data into that trial for a secondary exploratory analysis of biomarkers as mediators in the association between PAC use and blood pressure reductions.

In our exploratory analysis, there will be multiple testing bias due to utilizing a panel of 369 possible biomarker outcomes. With this size pilot cohort, it will be unlikely to achieve statistical significance if strict adjustments for multiple testing are undertaken. However, we will use the Holm procedure to evaluate for significance. Overall, these exploratory analyses will be hypothesis-generating and can help inform the design and sample size of future cohorts.

10.4 Sample Size

Power Analysis: While we do not expect substantial participant dropout over this short trial, our power analyses include adjustment for up to 10% dropout in each arm. Based on our meta-analysis of biomarkers[22], we estimate a 10% change in concentration of biomarkers with PAC use versus sham. [13 13 13 13] Using a basic multiple regression model incorporating age, BMI, and gender, along with our exposure (active PAC vs sham), we will have greater than 80% power to reject the null hypothesis of zero effect size at the significance level (alpha) of 0.05, with a sample size of 66. As such, our enrollment goal of 44 participants, limited by budgetary contraints, may provide sufficient power to detect significant changes in biomarker concentrations. However, in the event that we are not able to reach statistical significance with the changes we detect, these results will still be informative for developing future studies. We will include baseline-only data for participants who do not complete the protocol.

10.5 Measures to Minimize Bias

10.5.1 Enrollment/Randomization/Masking Procedures

We will block randomize participants for a net 1:1 allocation to active- versus sham-PAC arms. We will have one unblinded PI and one unblinded research coordinator; the remainder of the research staff will be blinded to each participant's treatment arm until the final visit (at which time, those in the sham PAC arm will receive HEPA filters). The allocation sequence file will be securely stored in an internal NYU OneDrive in a password protected file to which only the unblinded PI and unblinded research coordinator will have access.

PAC devices will be modified by unblinded research coordinator and/or unblinded PI; all PACs will have indicator lights covered, sham-PACs will have HEPA filters removed. PACs will be assigned a tracking ID number that will be printed on PACs, and PAC boxes. Blinded staff will only know the PAC ID number assigned to a participant but not the randomization of the device.

Unblinded research coordinator and PI will assess enrollment goals.

Blood samples will be assigned to PCR plate locations using a random number generator.

Data from PM monitors will be downloaded after participant has completed protocol. Research team members performing data processing of PM measurements will be blinded to PAC status of participant.

11 Source Documents and Access to Source Data/Documents

Source data include all study-related forms, including: completed surveys, checklists, interview transcripts, consent forms, air monitor documentation, and study personnel tracking sheets.
Access to study records will be limited to IRB-approved members of the study team. The investigator will permit study-related monitoring, audits, and inspections by the IRB/EC, the sponsor, government regulatory bodies, and University compliance and quality assurance groups of all study related documents.

Participation as an investigator in this study implies acceptance of potential inspection by government regulatory authorities and applicable University compliance and quality assurance offices.

12 Quality Assurance and Quality Control

QC procedures will be implemented beginning with the data entry system and data QC checks that will be run on the database will be generated. Any missing data or data anomalies will be communicated to the site(s) for clarification/resolution.

Following written SOPs, the monitors will verify that the clinical trial is conducted, and data are generated, documented (recorded), and reported in compliance with the protocol, GCP, and the applicable regulatory requirements (e.g., Good Laboratory Practices (GLP), Good Manufacturing Practices (GMP)).

The investigational site will provide direct access to all trial related sites, source data/documents, and reports for the purpose of monitoring and auditing by the sponsor, and inspection by local and regulatory authorities.

13 Ethics/Protection of Human Subjects

13.1 Ethical Standard

The investigator will ensure that this study is conducted in full conformity with Regulations for the Protection of Human Subjects of Research codified in 45 CFR Part 46.

13.2 Institutional Review Board

The protocol, informed consent form(s), recruitment materials, and all participant materials will be submitted to the IRB for review and approval. Approval of both the protocol and the consent form must be obtained before any participant is enrolled. Any amendment to the protocol will require review and approval by the IRB before the changes are implemented to the study. All changes to the consent form will be IRB approved; a determination will be made regarding whether previously consented participants need to be re-consented.

13.3 Informed Consent Process

13.3.1 Consent/Assent and Other Informational Documents Provided to Participants

Consent forms describing in detail the study intervention, study procedures, and risks are given to the participant and written documentation of informed consent is required prior to starting intervention. Forms

contain all regulatory consent text/content. The following consent materials are submitted with this protocol:

1. Consent for participation (including PAC use, pollution monitoring, BP monitoring and blood draws, and optional consent for blood collection/storage for future use)

13.3.2 Consent Procedures and Documentation

Informed consent is a process that is initiated prior to the individual's agreeing to participate in the study and continues throughout the individual's study participation. Extensive discussion of risks and possible benefits of participation will be provided to the participants and their families. Consent forms will be IRB-approved and the participant will be asked to read and review the document. The investigator will explain the research study to the participant and answer any questions that may arise. All participants will receive a verbal explanation in terms suited to their comprehension of the purposes, procedures, and potential risks of the study and of their rights as research participants. Participants will have the opportunity to carefully review the written consent form and ask questions prior to signing. The participants should have the opportunity to discuss the study with their surrogates or think about it prior to agreeing to participate. The participant will sign the informed consent document prior to any procedures being done specifically for the study. The participants may withdraw consent at any time throughout the course of the trial. A copy of the signed informed consent document will be given to the participants for their records. The rights and welfare of the participants will be protected by emphasizing to them that the quality of their medical care will not be adversely affected if they decline to participate in this study.

A copy of the signed informed consent document will be stored in the subject's research record. The consent process, including the name of the individual obtaining consent, will be thoroughly documented in the subject's research record. Any alteration to the standard consent process (e.g. use of a translator, consent from a legally authorized representative, consent document presented orally, etc.) and the justification for such alteration will likewise be documented.

13.4 Participant and Data Confidentiality

Information about study subjects will be kept confidential and managed according to the requirements of the Health Insurance Portability and Accountability Act of 1996 (HIPAA). Those regulations require a signed subject authorization informing the subject of the following:

- What protected health information (PHI) will be collected from subjects in this study
- Who will have access to that information and why
- Who will use or disclose that information
- The rights of a research subject to revoke their authorization for use of their PHI.

If a subject revokes authorization to collect or use PHI, the investigator, by regulation, retains the ability to use all information collected prior to the revocation of subject authorization. For subjects that have revoked authorization to collect or use PHI, attempts should be made to obtain permission to collect at least vital status (i.e. that the subject is alive) at the end of their scheduled study period.

This personal information will be retained, and all participants who enroll in the study will receive a unique study ID. Subsequent information will utilize this study ID instead of the participant's name, and the key linking the study IDs and personal information will be retained on the secured server, accessible only to the study PIs, data manager, and field staff communicating with those participants.

Participant confidentiality is strictly held in trust by the participating investigators, their staff, and the sponsor(s) and their agents. This confidentiality is extended to cover testing of biological samples and genetic tests in addition to the clinical information relating to participants. Therefore, the study protocol, documentation, data, and all other information generated will be held in strict confidence. No information concerning the study, or the data will be released to any unauthorized third party without prior written approval of the sponsor.

The study monitor, other authorized representatives of the sponsor, or representatives of the IRB may inspect all documents and records required to be maintained by the investigator, including but not limited to, medical records (office, clinic, or hospital) and pharmacy records for the participants in this study. The clinical study site will permit access to such records.

The study participant's contact information will be securely stored at each clinical site for internal use during the study. At the end of the study, all records will continue to be kept in a secure location for as long a period as dictated by local IRB and Institutional regulations.

Study participant research data, which is for purposes of statistical analysis and scientific reporting, will be transmitted to and stored at NYU Langone Medical Center. This will not include the participant's contact or identifying information. Rather, individual participants and their research data will be identified by a unique study identification number. The study data entry and study management systems used by clinical sites and by NYU Langone Medical Center research staff will be secured and password protected. At the end of the study, all study databases will be de-identified and archived at the NYU Langone Medical Center.

13.4.1 Research Use of Stored Human Samples, Specimens, or Data

Participants will have blood drawn, some of which will be used for biomarker measurement. The remaining portion of blood samples from participants will be stored in a secured laboratory in the Cardiology Division Science Building Laboratories.

- Intended Use: Samples and data collected under this protocol may be used to study additional pathways related to blood pressure and/or air pollution exposure.
- Storage: Access to stored samples will be limited to the study team only. Samples and data will be stored using codes assigned by the investigators. Data will be kept in password-protected computers. Only investigators will have access to the samples and data.
- Tracking: Data will be tracked using internal tracking datasheets stored in password protected file on NYU internal OneDrive. Study PIs will have password.
    - Disposition at the completion of the study: All samples will be stored in in a secured laboratory in the Cardiology Division Science Building Laboratories. Study participants who request destruction of samples will be notified of compliance with such request and all supporting details will be maintained for tracking.

13.5 Future Use of Stored Specimens

We will request participation in storage of blood samples collected for continued analyses of this study, for studies as new scientific discoveries are made, to allow for retesting if necessary and for future research related to cardiovascular risk. We will give subjects who participate in the study the option to opt into storage for future research as there are many unknowns about cardiovascular risk in patients with air pollution exposure, and being able to maintain the samples will allow us to build upon the knowledge we gain from this initial study. Patients will be informed of this during the consent process. Future research will not stray from the purpose of the original application, and optional participation in the storage of samples for future research will remain separated from the primary protocol. For optional future research, we will collect and additional ≈10 mL of blood at both the baseline and final blood draws.

Blood samples for future analysis will be assigned a unique code number and stored in a dedicated freezer storage by the PI without any identifying information other than a code number. The unique code number will not be based on any information that could be used to identify the subject (for example, social security number, initials, or birth date). We will use freezer-safe labels and store samples in freezers with emergency backup power. The master list linking names to code numbers will be kept in a locked file cabinet, separate from all research information, under double lock and key. Only the NYU PI will have access to the linking key between subject ID and subject identity. All confidential data will be stored with a unique code as an identifier and will be protected by a double electronic lock. All physical data will be kept under double physical lock. Only the PI and Co-Is will have access to the banked samples.

Samples will be stored for no more than 20 years. The specimens for future analysis may be analyzed upon completion of the study for RNA, microRNA, and DNA. The blood specimens may be analyzed upon completion of the study for RNA, microRNA, and DNA profiling. This may include detailed molecular profile study, describing genetic arrays of patterns associated with cardiometabolic risk. We will not be performing whole genome sequencing, only genetic testing as it relates to cardiovascular risk described above. Because there is no potential for clinically actionable genetic incidental findings because of this testing, genetic information will not be disclosed to anyone, including the participants and their physicians. Samples could also be used for testing other potential biomarkers in patient plasma and/or serum related to diabetes and CVD that may occur with exception of those which are already part of the protocol. Results of the future research will not be shared with the subjects or their physician, and will not become part of the participants' medical records.

In the future, other institutions, or future collaborators at or outside of NYU Medical Center may want to study a portion of these samples. If that happens, samples would be sent to other places so other people interested in studying these diseases and conditions could do that. The samples sent to other researchers will be de-identified without PHI. Samples will be stored for no more than 20 years following completion of the study. At the end of the 20-year period, all samples will be destroyed.

Subjects can request for withdrawal of samples at any time by contacting PI Dr. Newman in writing. His mailing address is NYU Translational Research Building, 227 E. 30th St, Suite 853, NEW YORK 10016. Withdrawing Authorization only affects uses and sharing of information after written request has been received, and subject may not withdraw his/her Authorization for uses or disclosures that have previously been made or must continue to complete analyses or report data from the research.

14 Data Handling and Record Keeping

14.1 Data Collection and Management Responsibilities

Data collection is the responsibility of the study staff at the site under the supervision of the site PIs. The investigator is responsible for ensuring the accuracy, completeness, legibility, and timeliness of the data reported.

All source documents should be completed in a neat, legible manner to ensure accurate interpretation of data. Black ink is required to ensure clarity of reproduced copies. When making changes or corrections, cross out the original entry with a single line, and initial and date the change. DO NOT ERASE, OVERWRITE, OR USE CORRECTION FLUID OR TAPE ON THE ORIGINAL.

Copies of the electronic CRF (eCRF) will be provided for use as source documents and maintained for recording data for each participant enrolled in the study. Data reported in the eCRF derived from source documents should be consistent with the source documents or the discrepancies should be explained and captured in a progress note and maintained in the participant's official electronic study record.

Clinical data (including AEs, concomitant medications, and expected adverse reactions data) and clinical laboratory data will be entered into RedCAP. The data system includes password protection and internal quality checks, such as automatic range checks, to identify data that appear inconsistent, incomplete, or inaccurate. Clinical data will be entered directly from the source documents.

For questionnaires confirming inclusion/exclusion criteria, data will be entered on an iPad with a data plan, provided by NYUMC, and stored using REDCap. No identifiable information will be entered or will be able to be accessed by field staff. In addition, iPads with be programmed to limit access to only REDCap prior to data collection. Back-up paper forms will be used in the event of technology issues. After data cleaning and quality assurance procedures are completed, pertinent sets of data will be converted into SAS format for statistical analyses. Only authorized research staff will have access to the files.

14.2 Study Records Retention

Study documents will be retained for at least 5 years after final reporting/publication. No records will be destroyed without the written consent of the sponsor.

14.3 Protocol Deviations

All deviations in protocol will be addressed in study source documents, reported to the NYULMC IRB.

14.4 Publication and Data Sharing Policy

This study will comply with the NIH Public Access Policy, which ensures that the public has access to the published results of NIH funded research. It requires scientists to submit final peer-reviewed journal manuscripts that arise from NIH funds to the digital archive PubMed Central upon acceptance for publication.

The International Committee of Medical Journal Editors (ICMJE) member journals have adopted a clinical trials registration policy as a condition for publication. The ICMJE defines a clinical trial as any research project that prospectively assigns human subjects to intervention or concurrent comparison or control groups to study the cause-and-effect relationship between a medical intervention and a health outcome. Medical interventions include drugs, surgical procedures, devices, behavioral treatments, process-of-care changes, and the like. Health outcomes include any biomedical or health-related measures obtained in patients or participants, including pharmacokinetic measures and adverse events. The ICMJE policy, and the Section 801 of the Food and Drug Administration Amendments Act of 2007, requires that all clinical trials be registered in a public trials registry such as ClinicalTrials.gov, which is sponsored by the National Library of Medicine. Other biomedical journals are considering adopting similar policies. For interventional clinical trials performed under NIH IC grants and cooperative agreements, it is the grantee’s responsibility to register the trial in an acceptable registry, so the research results may be considered for publication in ICMJE member journals. The ICMJE does not review specific studies to determine whether registration is necessary; instead, the committee recommends that researchers who have questions about the need to register err on the side of registration or consult the editorial office of the journal in which they wish to publish.

FDAAA mandates that a "responsible party" (i.e., the sponsor or designated principal investigator) register and report results of certain "applicable clinical trials":

- Trials of Drugs and Biologics: Controlled, clinical investigations, other than Phase I investigations of a product subject to FDA regulation;
- Trials of Devices: Controlled trials with health outcomes of a product subject to FDA regulation (other than small feasibility studies) and pediatric postmarket surveillance studies.
- NIH grantees must take specific steps to ensure compliance with NIH implementation of FDAAA.

15 Study Finances

15.1 Funding Source

This study is financed through a pilot grant from the NYU CTSI under grant number NIH/NCATS UL1TR001445

15.2 Costs to the Participant

No costs will incur as a result of participating in the study.

15.3 Participant Reimbursements or Payments

Individual participants will be remunerated for blood draws as follows: Participants will receive $25 after the baseline blood draw. Upon completing the study, participants will be allowed to keep the air cleaner,

and will be given a true filter if assigned to the “sham” group; participants will also keep the blood pressure cuff. Participants will receive $75 after all other equipment is returned (electricity monitor, air pollution monitor and blood pressure data hub) and the final blood draw is completed.

16 Study Administration

16.1 Study Leadership

This protocol describes a co-PI collaborative pilot project bringing together a well-established investigator and a post-doctoral research fellow with complementary skills and expertise, a shared commitment to the objectives of this study and a history of collaboration.

The PI of the study is Dr. Jonathan Newman, the Eugene Braunwald M.D. Assistant Professor of Cardiology (Medicine). Dr. Newman has a Master’s in Public Health in clinical research methodology and has substantial experience leading clinical trials, including the ISCHEMIA trial, the TACT2 trial, NYU (REPAIR). Dr. Newman has led recent investigations into the environmental effects of air pollution exposure, including a recent NIH/CDC/EPA white paper defining strategies to reduce the cardiopulmonary impact of particulate air pollution. The Co-PI of the study is Dr. Sharine Wittkopp, MD, PhD, Cardiology research fellow. Dr. Wittkopp has a Master’s in Biomedical Science on mitochondrial biochemistry and oxidative stress, with experience studying inflammation and air pollution exposure in human subjects cohorts in the CHAPS study of the Los Angeles air basin.

Drs. Newman and Wittkopp recently collaborated with Co-investigators Drs. Thorpe and Gordon on a feasibility study on the use of PACs and home BP monitoring in a community cohort. The team’s work on this recent feasibility study has informed the design of the current pilot study presented herein.

17 Conflict of Interest Policy

The independence of this study from any actual or perceived influence is critical. Therefore any actual conflict of interest of persons who have a role in the design, conduct, analysis, publication, or any aspect of this study will be disclosed and managed. Furthermore, persons who have a perceived conflict of interest will be required to have such conflicts managed in a way that is appropriate to their participation in the study. The study leadership has established policies and procedures for all study group members to disclose all conflicts of interest and will establish a mechanism for the management of all reported dualities of interest.

Any investigator who has a conflict of interest with this study (patent ownership, royalties, or financial gain greater than the minimum allowable by their institution, etc.) must have the conflict reviewed by the NYU Langone Conflict of Interest Committee with a Committee-sanctioned conflict management plan that has been reviewed and approved by the study sponsor prior to participation in this study. All NYULMC investigators will follow the applicable conflict of interest policies.

18 References

1. Yang BY, Qian Z, Howard SW, et al. Global association between ambient air pollution and blood pressure: A systematic review and meta-analysis. *Environ Pollut*. Apr 2018;235:576-588. doi:10.1016/j.envpol.2018.01.001
2. Liang R, Zhang B, Zhao X, Ruan Y, Lian H, Fan Z. Effect of exposure to PM2.5 on blood pressure: a systematic review and meta-analysis. *J Hypertens*. Nov 2014;32(11):2130-40; discussion 2141. doi:10.1097/HJH.0000000000000342
3. Brook RD, Brook JR, Urch B, Vincent R, Rajagopalan S, Silverman F. Inhalation of fine particulate air pollution and ozone causes acute arterial vasoconstriction in healthy adults. *Circulation*. Apr 2 2002;105(13):1534-6. doi:10.1161/01.cir.0000013838.94747.64

4. Brook RD, Urch B, Dvonch JT, et al. Insights into the mechanisms and mediators of the effects of air pollution exposure on blood pressure and vascular function in healthy humans. *Hypertension*. Sep 2009;54(3):659-67. doi:10.1161/HYPERTENSIONAHA.109.130237
5. Wittkopp S, Staimer N, Tjoa T, et al. Nrf2-related gene expression and exposure to traffic-related air pollution in elderly subjects with cardiovascular disease: An exploratory panel study. *J Expo Sci Environ Epidemiol*. Mar-Apr 2016;26(2):141-9. doi:10.1038/jes.2014.84
6. Pope CA, 3rd, Hansen ML, Long RW, et al. Ambient particulate air pollution, heart rate variability, and blood markers of inflammation in a panel of elderly subjects. *Environ Health Perspect*. Mar 2004;112(3):339-45. doi:10.1289/ehp.6588
7. Calderon-Garciduenas L, Villarreal-Calderon R, Valencia-Salazar G, et al. Systemic inflammation, endothelial dysfunction, and activation in clinically healthy children exposed to air pollutants. *Inhal Toxicol*. Mar 2008;20(5):499-506. doi:10.1080/08958370701864797
8. Liu C, Cai J, Qiao L, et al. The Acute Effects of Fine Particulate Matter Constituents on Blood Inflammation and Coagulation. *Environ Sci Technol*. Jul 18 2017;51(14):8128-8137. doi:10.1021/acs.est.7b00312
9. Tornqvist H, Mills NL, Gonzalez M, et al. Persistent endothelial dysfunction in humans after diesel exhaust inhalation. *Am J Respir Crit Care Med*. Aug 15 2007;176(4):395-400. doi:10.1164/rccm.200606-872OC
10. Lamb FS, Choi H, Miller MR, Stark RJ. TNFalpha and Reactive Oxygen Signaling in Vascular Smooth Muscle Cells in Hypertension and Atherosclerosis. *Am J Hypertens*. Oct 21 2020;33(10):902-913. doi:10.1093/ajh/hpaa089
11. Rich DQ, Kipen HM, Huang W, et al. Association between changes in air pollution levels during the Beijing Olympics and biomarkers of inflammation and thrombosis in healthy young adults. *JAMA*. May 16 2012;307(19):2068-78. doi:10.1001/jama.2012.3488
12. Walzer D, Gordon T, Thorpe L, et al. Effects of Home Particulate Air Filtration on Blood Pressure: A Systematic Review. *Hypertension*. Jul 2020;76(1):44-50. doi:10.1161/HYPERTENSIONAHA.119.14456
13. Wittkopp S, Walzer D, Thorpe L, et al. Meta-Analysis of Residental Portable Air Cleaner Use and Circulating Inflammatory Biomarkers. Abstract presented at: AHA Scientific Sessions; 11/13/2021 2021; Boston, MA. Session Exercise, Diet and Lifestyle Influences on CVD.
14. Riggs DW, Zafar N, Krishnasamy S, et al. Exposure to airborne fine particulate matter is associated with impaired endothelial function and biomarkers of oxidative stress and inflammation. *Environ Res*. Jan 2020;180:108890. doi:10.1016/j.envres.2019.108890
15. Macchi C, Ferri N, Favero C, et al. Long-term exposure to air pollution raises circulating levels of proprotein convertase subtilisin/kexin type 9 in obese individuals. *Eur J Prev Cardiol*. Apr 2019;26(6):578-588. doi:10.1177/2047487318815320
16. Zhao Y, Xue L, Chen Q, et al. Cardiorespiratory responses to fine particles during ambient PM2.5 pollution waves: Findings from a randomized crossover trial in young healthy adults. *Environ Int*. Jun 2020;139:105590. doi:10.1016/j.envint.2020.105590
17. Vieira JL, Guimaraes GV, de Andre PA, Cruz FD, Saldiva PH, Bocchi EA. Respiratory Filter Reduces the Cardiovascular Effects Associated With Diesel Exhaust Exposure: A Randomized, Prospective, Double-Blind, Controlled Study of Heart Failure: The FILTER-HF Trial. *JACC Heart Fail*. Jan 2016;4(1):55-64. doi:10.1016/j.jchf.2015.07.018
18. deFilippi C, Lo J, Christenson R, et al. Novel mediators of statin effects on plaque in HIV: a proteomics approach. *AIDS*. Apr 24 2018;32(7):867-876. doi:10.1097/QAD.0000000000001762
19. Alonso A, Chen LY, Rudser KD, Norby FL, Rooney MR, Lutsey PL. Effect of Magnesium Supplementation on Circulating Biomarkers of Cardiovascular Disease. *Nutrients*. Jun 6 2020;12(6)doi:10.3390/nu12061697
20. Bullo M, Garcia-Lorda P, Megias I, Salas-Salvado J. Systemic inflammation, adipose tissue tumor necrosis factor, and leptin expression. *Obes Res*. Apr 2003;11(4):525-31. doi:10.1038/oby.2003.74
21. Franceschi C, Campisi J. Chronic inflammation (inflammaging) and its potential contribution to age-associated diseases. *J Gerontol A Biol Sci Med Sci*. Jun 2014;69 Suppl 1:S4-9. doi:10.1093/gerona/glu057
22. Wittkopp S, Walzer D, Thorpe L, et al. Portable Air Cleaner Use and Biomarkers of Inflammation: A Systematic Review and Meta-Analysis. *American Heart Journal*. in review 2022;

19 Attachments

These documents are relevant to the protocol, but they are not considered part of the protocol. They are stored and modified separately. As such, modifications to these documents do not require protocol amendments.

- Attachment A: Comorbidity and medication questionnaire
- Attachment B: Instructions for BP recording
- Attachment C: Instructions for Equipment Setup

Attachment A : Air Filter Pilot Study Questionnaire

Pre-Intervention Questionnaire:

Height and Weight Measurements (self-reported measures):
Enter the respondent's weight (lbs.) here. Weight to the nearest 0.2 lb.: XXX
Enter the respondent's height (inches) here. Height to the nearest 0.5 in.: XXX

1. How long have you had high blood pressure?
    a. 0-2 years
    b. 2-5 years
    c. 5-10 years
    d. >10 years
    e. Don't know/refused

2. How long as it been since you last had your blood pressure taken by a doctor or other health professional? Was it…
    a. Within the past month?
    b. 1-6 months ago?
    c. 6-12 months ago?
    d. Over 1 year ago?
    e. Don't know/refused

3. Have you ever been told to take prescription medicine because of high blood pressure?
    a. Yes
    b. No
    c. Don't know/refused

4. Have you been told to take or continue taking prescription medicine because of high blood pressure/hypertension in the past 12 months?
    a. Yes
    b. No
    c. Don't know/refused

5. *[If yes to Q3/Q4]:* Are you now taking prescription medicine?
    a. Yes
    b. No
    c. Don't know/refused

6. Have you ever been told you have an inflammatory disorder such as rheumatoid arthritis, inflammatory bowel disease, cancer?
    a. Yes
    b. No
    c. Don't know/refused

7. If you take medications to lower your blood pressure, have any of these medications changed in the last month?
    a. Yes
    b. No
    c. Don't know/refused

8. In the past month, how often have you checked your blood pressure at home?
    a. Never
    b. Less than weekly
    c. Weekly

d. More than weekly

9. Have you ever had, or been told you had, a heart attack?
    a. Yes
    b. No
    c. Don't know/refused

10. Have you ever had, or been told you had, a stroke?
    a. Yes
    b. No
    c. Don't know/refused

11. How concerned are you about the effects of air pollution on your health?
    a. Extremely concerned
    b. Very concerned
    c. Somewhat concerned
    d. Mildly concerned
    e. Not concerned

(On Day 0, trained field staff will enter participant's home, and place field equipment inside their home, providing detailed demonstration on how to measure blood pressure. Trained field staff will also manually record participant's height and weight (lbs.) using a digital scale)

Post-Intervention Questionnaire:

Now that you have an air filter, I have some questions for you:

1. How much time each day did you spend checking your BP for the study?
    a. 2-5 minutes
    b. 5-10 minutes
    c. 10-15 minutes

2. On average, how much time did you run the filter each day?
    a. I kept it on for 24 hours a day
    b. I ran it for most of the day (18-24 hours)
    c. I ran it for more than half of the day (12-18 hours)
    d. I ran it for part of the day (6-12 hours)
    e. I ran it for a few hours (0-6 hours)

3. How much did you notice noise from the filter?
    a. Extremely noisy
    b. Very noisy
    c. Somewhat noisy
    d. Minimal noise
    e. I didn't hear it at all

4. How likely is it that you will buy replacement filters in the future? [*Likert scale*]
    a. Very likely
    b. Somewhat likely
    c. Not very likely
    d. Not likely at all

5. *[If less than very likely….]* What are some of the reasons why you might not buy replacement air filters (check all that apply)?
    a. Too costly
    b. Too inconvenient
    c. Don't know where to buy them
    d. Too noisy

e. Not sure they are effective
f. Other: _____

6. During the study period did you have a change in your blood pressure medication?
    a. Yes
    b. No
    c. Don't know/refused

7. How concerned are you about the effects of air pollution on your health?
    a. Very concerned
    b. Somewhat concerned
    c. Not very concerned
    d. Not concerned at all

8. Would you be interested in continuing to participant in this study and check your BP twice daily for 3 months?
    a. Yes
    b. No
    c. Don't know/refused

9. If this study continued for 6 months, would you be interested in continuing to participate and check your BP daily?
    a. Yes
    b. No
    c. Don't know/refused

Attachment B: Participant Instructions for Measuring Blood Pressure

Checking your blood pressure at home is an important part of this study and helps you know when your blood pressure is high. Here is a quick guide on how to check your blood pressure at home.

1. Please take two blood pressure readings 1 minute apart in the morning and two readings 1 minute apart in the evening for every day for 2 weeks (for a total of 4 per day, and 64 readings over the 2 weeks).

2. Make sure the air plug of the blood pressure cuff is placed tight into the machine before taking your blood pressure.

3. Remove tight-fitting clothing from the left upper arm.

4. Sit in a chair with your feet flat on the floor. Put your left arm on a table or armrest so the cuff is about the same height as your heart. Sit for at least 5 minutes prior to taking your first blood pressure reading.

5. Place the blood pressure cuff on the left upper arm so that the white stripe is on the inside of the arm. Place the bottom of the blood pressure cuff about 1/2 inch above the elbow. Make sure that the air tube is pointed downward – it should run down the inside of your forearm and be in line with your middle finger (with the palm up). **See next page for a diagram.**

6. Make sure that you are comfortable, and your left arm is not tense. Keep your left hand open.

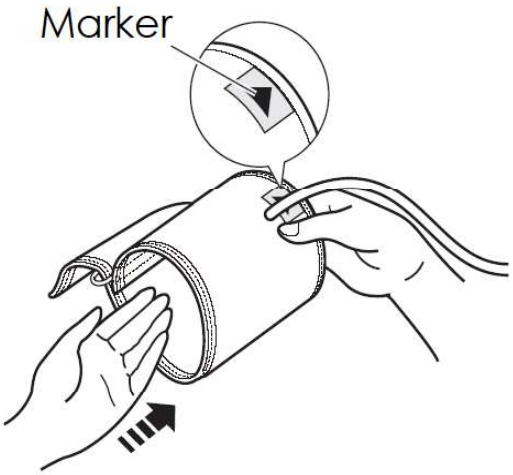
7. Press the Start/Stop button once with your other hand to take a measurement. You should feel the blood pressure cuff tighten when the blood pressure is being taken. Do not move or talk, and stay relaxed. Wait until the cuff deflates and your blood pressure is showing on the display. Once that has occurred, remove the blood pressure cuff.

   You can use this table to keep track of whether or not you have measured your blood pressure at these times (Example: place 2X's under Day 1 AM after you've collected your two morning readings)

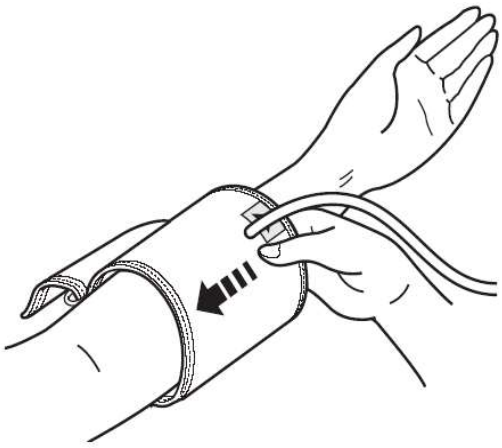
Week 1 (date range: / / to / /)							
	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7
AM							
PM							
Week 2 (date range: / / to / /)							
	Day 8	Day 9	Day 10	Day 11	Day 12	Day 13	Day 14
AM							
PM							
Week 3 (date range: / / to / /)							
	Day 8	Day 9	Day 10	Day 11	Day 12	Day 13	Day 14
AM							
PM							
Week 4 (date range: / / to / /)							
	Day 8	Day 9	Day 10	Day 11	Day 12	Day 13	Day 14
AM							
PM							

HOW TO APPLY CUFF

1 Put your arm through the cuff loop.



2 Insert your arm as indicated by the marker.

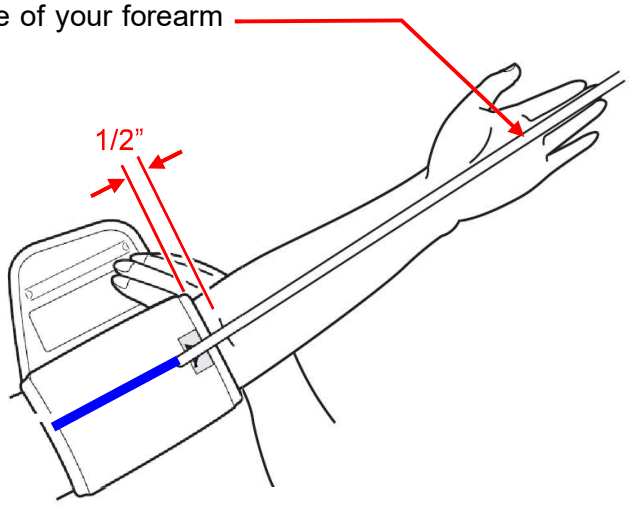


3 Position the arm correctly.

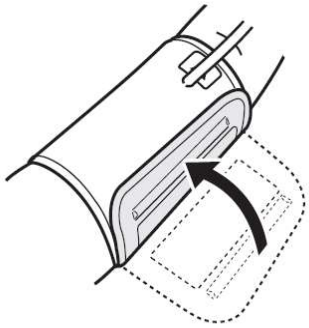
a) The air tube should run down the inside of your forearm and be in line with your middle finger.

b) The bottom of the cuff should be approximately 1/2” above your elbow.

c) Make sure that the blue stripe is on the inside of the arm and aligned with the middle finger (with the palm up).



4 Close the fabric (Velcro) fastener FIRMLY.

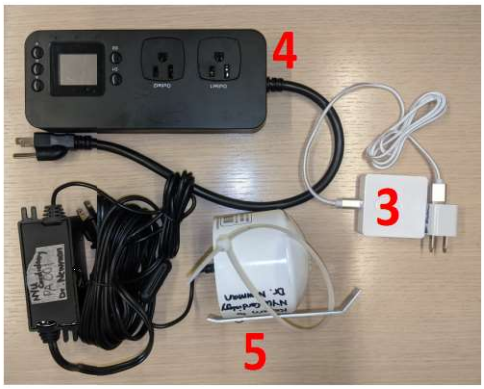


Attachment C: Participant Instructions for Equipment Setup

Equipment Setup Instructions:

You have received:

1. Blood pressure cuff
2. Home Air Cleaner
3. Electricity monitor
4. Data Hub & cord
5. Air Pollution monitor & cord



- Plug the electricity monitor (4) into an outlet in your bedroom/sleeping area.
- Plug the Air Cleaner (2) into outlet number 1 on the electricity monitor.
- Turn the Air cleaner on to level 1. (You will leave it like this for the whole study.)
- Hang the air pollution monitor (5) to a wall on the opposite side of the same room – use the included command hook to secure it to a wall about 4 feet above an outlet.
- Plug in the air pollution monitor.
- Plug in the data hub (3) within 10 feet of where you will check your blood pressure (1). Please see separate instructions for use.

Bring to your final visit:

3. Electricity monitor
4. Data Hub & cord
5. Air Pollution monitor & cord