Document: Study Protocol and Statistical Analysis Plan

Study Title: Angiotensin II Receptor Inhibition to Improve Microvascular Function in Women Who Have Had Preeclampsia

NCT# 04632589

Document Date(s):

Updated and approved by IRB 7/19/2024

Downloaded 6/2/2025

Losartan for Vascular Function after Preeclampsia

PI: Anna Reid-Stanhewicz
IRB ID #: 202006148

Project Details

I. Project Introduction

I.1 ***Project to be reviewed by:***
IRB-01

I.2 ***Project Title:***
Angiotensin II Receptor Inhibition to Improve Microvascular Function in Women who have had Preeclampsia

I.3 ***Short Title (optional):***
Losartan for Vascular Function after Preeclampsia

I.4 ***Provide a short summary of the purpose and procedures of the study proposed in this IRB application.***

- ***DO NOT include information on studies not proposed in this application.***
- ***Use LAY terminology only. This must be easily understandable by IRB community members and nonscientists.***
- ***DO NOT cut and paste technical abstracts from funding applications that may not be understood by a general audience.***

Women who develop preeclampsia during pregnancy are more likely to develop and die of cardiovascular disease later in life, even if they are otherwise healthy. The reason why this occurs is unclear but may be related to blood vessel damage and increased inflammation that occurs during the preeclamptic pregnancy and persists postpartum. The purpose of this investigation is to determine the mechanisms contributing to this lasting blood vessel damage and chronic inflammation, and to test whether taking a medication that blocks angiotensin II receptors (losartan) decrease these negative effects in women who have had preeclampsia. Identification of these mechanisms and treatment strategies may lead to better clinical management of cardiovascular disease risk in these women.

In this study we are recruiting women who have had preeclampsia in the past 5 years. We use the blood vessels in the skin as a representative vascular bed. Using a minimally invasive technique (intradermal microdialysis for the local delivery of pharmaceutical agents) we examine the blood vessels in a nickle-sized area of the skin in women who have had preeclampsia. We make these measurements after the subjects take a placebo and after they take losartan (an angiotensin II receptor blocker) to test whether this treatment improves vascular function in these women. Because losartan can have dangerous effects on a fetus, women enrolled in this study will be required to use an effective form of birth control [hormonal patch, implant or pill; intrauterine device; consistent use of barrier contraceptive; or prior medical procedure to prevent pregnancy (i.e. tubal ligation or vasectomy in current partner)]. As a compliment to these measurements, we also draw blood from the subjects and isolate the inflammatory cells to test how sensitive their inflammatory responses are following the placebo and the losartan treatment.

A modification was submitted changing the eligibility requirements from having preeclampsia within the past 2 years, to within the past 5 years. This modification was made to help with recruitment for the study. Women who are breastfeeding are excluded from this study. Since many women are breastfeeding during those first 2 years after delivery, it reduced those that were eligible for the study. We changed it to delivering within the past 5 years to match the other preeclampsia studies in our lab (IRB #202203433, 202303202, 202303799).

I.5 ***Specify your research question(s), study aims or hypotheses (do not indicate "see protocol")***
AIM: To determine the effect of systemic AT1R inhibition (oral losartan) on in vivo and in vitro measures of inflammation and endothelial function in women who have had preeclampsia (PE) . Utilizing a chronic (6 week) losartan intervention coupled with intradermal microdialysis and assessment of the inflammatory response to ang II stimulation in peripheral blood mononuclear cells (PBMC), we will test the hypotheses that therapeutic doses of the AT1R inhibitor augment endothelium-dependent dilation and reduce inflammation in PE.

HYPOTHESES
1: Chronic oral losartan therapy will augment endothelium-dependent dilation in PE by increasing nitric oxide-dependent dilation.
2: Chronic oral losartan therapy will attenuate exaggerated ang II sensitivity in PE.
3: Chronic oral losartan therapy will reduce circulating inflammatory cytokines, and attenuate the exaggerated inflammatory response to ang II stimulation in PBMCs from PE.

NOTE: This IRB protocol encompasses the projects of aim 3 in the attached NIH K99/R00 award. Aim 2 has been completed and aim 1 is approved under HawIRB #201909818.

I.6 ***Background and significance and/or Preliminary studies related to this project.***
(do not indicate "see protocol")
Angiotensin II Sensitivity Contributes to Endothelial Dysfunction Following Preeclampsia: Women with a history of preeclampsia have an exaggerated presser response to systemic infusion of Ang II. Similarly, compelling in vivo pilot data from our lab demonstrates that this increased sensitivity to Ang II is present in the microcirculation and contributes to endothelial dysfunction postpartum in formerly preeclamptic patients. Ang II mediates several events of the inflammatory process including activation of the vascular endothelium; and AT1R activation plays a large role in the pathophysiology of concurrent inflammatory reactions through this direct action on local vascular cells. Inhibition of Ang II, either through ACE inhibition or AT1-receptor antagonism, reduces tissues inflammation and oxidative injury. Reciprocally, inhibition of inflammatory mediators such as IL-17 producing T cells blunts the pressor response to systemic Ang II infusion in animal models of hypertension. Collectively, increased sensitivity to Ang II coupled with the role of Ang II signaling in chronic inflammation, suggests that these factors are likely reciprocal, and contribute to microvascular dysfunction and elevated CVD risk in women who have had preeclampsia.

AT1 Receptor Inhibition Is a Viable Intervention Strategy to Mitigate CVD Risk Following Preeclampsia: The clinical symptoms of preeclampsia (high blood pressure, proteinuria, edema, etc.) typically resolve within 12 weeks post-partum. However, despite this absence of

clinical CVD, women who have had preeclampsia demonstrate attenuated endothelium-dependent dilation, augmented ang II sensitivity, and elevated inflammation, indicating the need for early intervention. Aside from its primary antihypertensive effects, AT1R inhibition prevents the production of inflammatory mediators and reduces biomarkers of inflammation in populations with known CVD. Similarly, chronic AT1R inhibition improves endothelium-dependent dilation in patients with vascular dysfunction independent of reduction in blood pressure. Given that women who have had preeclampsia have a greater sensitivity to ang II, AT1R inhibition may be a mechanistically specific intervention strategy to reduce inflammation and ameliorate endothelial dysfunction in this population. However, to date, no human studies have mechanistically examined this specific pharmacotherapy in women who have had preeclampsia.

I.7 ***Literature cited / references (if attaching a grant or protocol enter N/A).***
NA

II. Research Team

II.1 ***Principal Investigator***

Name	E-mail	College
Anna Reid-Stanhewicz	anna-stanhewicz@uiowa.edu	College Lib Arts and Sciences

II.2 ***Team Members***

UI Team Members

Name	E-mail	College	Contact	Key Prsn	UI COI	VAMC COI	Consent Process Involvement	Deactivated
Anna Reid-Stanhewicz, PHD	anna-stanhewicz@uiowa.edu	College Lib Arts and Sciences	Yes	Yes	No		Yes	No
Adam Corkery, PHD	adam-corkery@uiowa.edu	College Lib Arts and Sciences	No	Yes	No		Yes	No
Vincenzo Dimarco, High School	vincenzo-dimarco@uiowa.edu	College of Liberal Arts and Sciences	No	No	No		No	No
Alyssa Engels, High School	alyssa-engels@uiowa.edu	College of Liberal Arts and Sciences	No	No	No		No	No
Ashley Faber, High School	ashley-m-faber@uiowa.edu		No	No	No		No	No
Katharine Geasland, BSN	katharine-geasland@uiowa.edu	Inst Clinical & Translational	No	No	No		No	No
Claire Goebel, BA	claire-goebel@uiowa.edu	College Lib Arts and Sciences	No	No	No		Yes	No
Diana Jalal, MD	diana-jalal@uiowa.edu	Carver College of Medicine	No	Yes	No		No	No
Isabel Klimowicz, High School	isabel-klimowicz@uiowa.edu	College of Liberal Arts and Sciences	No	No	No		No	No
Ruda Lee, MS	ruda-lee@uiowa.edu	Graduate College	No	No	No		Yes	No
Maddie Marino, High School	madalyn-marino@uiowa.edu	College of Liberal Arts and Sciences	No	No	No		No	No
Grace Maurer, MS	grace-maurer@uiowa.edu	Graduate College	No	No	No		Yes	No
Leah Patton, High School	leah-patton@uiowa.edu	College of Liberal Arts and Sciences	No	No	No		No	No
Gary Pierce, PHD, MS	gary-pierce@uiowa.edu	College Lib Arts and Sciences	No	Yes	No		No	No
Bomi Ryang, BS	bomi-ryang@uiowa.edu	College of Pharmacy	No	No	No		No	No
Kelsey Schwartz, BS	kelsey-schwartz@uiowa.edu	Carver College of Medicine	No	No	No		Yes	No
Nikolas Soofi, High School	nikolas-soofi@uiowa.edu	College of Liberal Arts and Sciences	No	No	No		No	No
Amy Marie Stroud, MSN	amy-stroud@uiowa.edu	Carver College of Medicine	No	Yes	No		No	No
Olivia Taeger, High School	olivia-taeger@uiowa.edu	College of Liberal Arts and Sciences	No	No	No		No	No
Lizzy Wetzel, High School	elizabeth-wetzel@uiowa.edu	UIHC University Hospital	No	No	No		No	No
Ellie Wilson, High School	ellie-wilson@uiowa.edu	College of Liberal Arts and Sciences	No	No	No		No	No

Non-UI Team Members

Name Institution Location FWA Role DHHS Contact Key Prsn UI COI VAMC COI Consent Process Involvement Email

Nothing found to display.

II.3 ***The Principal Investigator of this study is:***
Faculty

II.5 ***Select research team member who is the primary contact for study participants.***
Claire Goebel

II.6 ***Identify the key personnel. The system will automatically designate the PI and all faculty members on the project as "key personnel." For information about other team members who should be designated as "key personnel" please click on the help information.***

Name	Is Key Personnel
Anna Reid-Stanhewicz, PHD	Yes
Adam Corkery, PHD	Yes
Vincenzo Dimarco, High School	No
Alyssa Engels, High School	No
Ashley Faber, High School	No
Katharine Geasland, BSN	No
Claire Goebel, BA	No
Diana Jalal, MD	Yes
Isabel Klimowicz, High School	No
Ruda Lee, MS	No
Maddie Marino, High School	No
Grace Maurer, MS	No
Leah Patton, High School	No
Gary Pierce, PHD, MS	Yes
Bomi Ryang, BS	No
Kelsey Schwartz, BS	No
Nikolas Soofi, High School	No
Amy Marie Stroud, MSN	Yes
Olivia Taeger, High School	No
Lizzy Wetzel, High School	No
Ellie Wilson, High School	No

III. Funding/Other Support

III.1 ***Select all sources of funding and/or support that will be applied to this research (select all that apply):***

- ☐ The funding award (contract/grant) has been routed through the Division of Sponsored Programs (DSP)
- ☐ University of Iowa Center for Advancement (UICA)
- ☐ Howard Hughes Medical Institute (private foundation)
- ☐ Fellowship Award
- ☐ In Kind Donations
- ☐ PI Discretionary
- ☑ Departmental
- ☐ UI Institutional Grant/Award
- ☐ PI Personal Funds (students only)
- ☐ No Funding (students only)

III.2 ***Funding Sources***

Source Entered as Text	DSP Link	Type	Source	Project Title	Na
Source is entered as text no		Federal Agency	US Department of Health & Human Services, National Institutes of Health	Role of Angiotensin II and Chronic Inflammation in Persistent Microvascular Dysfunction Following Preeclamptic Pregnancy	An Sta

* new source name

III.3 ***Does any member of the research team have a financial conflict of interest related to this project according to the Conflict of Interest in Research policy? If yes, please indicate which members below.***

Name	Has Conflict of Interest
Anna Reid-Stanhewicz, PHD	No
Adam Corkery, PHD	No
Vincenzo Dimarco, High School	No

Alyssa Engels, High School	No
Ashley Faber, High School	No
Katharine Geasland, BSN	No
Claire Goebel, BA	No
Diana Jalal, MD	No
Isabel Klimowicz, High School	No
Ruda Lee, MS	No
Maddie Marino, High School	No
Grace Maurer, MS	No
Leah Patton, High School	No
Gary Pierce, PHD, MS	No
Bomi Ryang, BS	No
Kelsey Schwartz, BS	No
Nikolas Soofi, High School	No
Amy Marie Stroud, MSN	No
Olivia Taeger, High School	No
Lizzy Wetzel, High School	No
Ellie Wilson, High School	No

IV. Project Type

IV.1 ***Do you want the IRB to give this project***
Regular (expedited or full board) review

IV.2 ***Enter the date you will be ready to begin screening subjects/collecting data for this project. (If you do not have a specified date, add "upon IRB approval")***
upon IRB approval

IV.3 ***Are you requesting a waiver of informed consent/authorization (subjects will not be given any oral or written information about the study)?***
No

V. Other Committee Review

V.1 ***Does this project involve any substance ingested, injected, or applied to the body?***

- ***Do not answer yes, if the involvement includes a device, wire, or instrument***

Yes

V.1.a ***What is/are the substance(s):***
Note: All of the following research agents used with microdialysis have been approved by the FDA for this study under IND# 124,294. A modification to this IND has been submitted to include oral losartan.

Acetylcholine, Powder
Angiotensin II, Powder
NG-nitro-L-arginine methyl ester, Powder
Norepinephrine, Powder
Sodium Nitroprusside, Powder
Lactated Ringer's, Liquid

Losartan, Tablet
Placebo, Tablet

V.1.b ***Are any of these substances defined as a Schedule I - V Controlled Substance?***
No

V.2 ***Are any contrast agents used for any purpose in this study?***
No

V.4 ***Are all drugs or substances in this study being used within the FDA approved population (i.e., children, adults)?***
Yes

V.5 ***Are all drugs or substances in this study being used within the FDA approved indication (i.e., disease, condition)?***
No

V.6 ***Are all drugs or substances in this study being used within the FDA approved dose?***
No

V.7 ***Are all drugs or substances in this study being used within the FDA approved route of administration?***
No

V.8 ***Drugs used in study that are not FDA approved for the population, indication, dose, or route of administration***

Ng-nitro-L-arginine-methyl ester (L-NAME) ()

Name of Sponsor	Anna Stanhewicz
Investigator's Brochure Version	NA
Investigator's Brochure Date	NA
Who is supplying the drug	Tocris
Who is dispensing the drug	PI/Research Team
Where will the drug be stored	118 PSRB
IND#	124,294
Dose	<0.01mg
Route of administration	intradermal microdialysis

losartan ()

Name of Sponsor	Anna Stanhewicz
Investigator's Brochure Version	NA
Investigator's Brochure Date	NA
Who is supplying the drug	NuCara Pharmacy
Who is dispensing the drug	PI/Research Team
Where will the drug be stored	NA - individual prescriptions obtained from NuCara, dispensed to participant immediately
IND#	124,294
Dose	50mg/day for 6 weeks
Route of administration	oral

lactated Ringer's solution ()

Name of Sponsor	Anna Stanhewicz
Investigator's Brochure Version	NA
Investigator's Brochure Date	NA
Who is supplying the drug	VWR
Who is dispensing the drug	PI/Research Team
Where will the drug be stored	118 PSRB
IND#	124,294
Dose	NA
Route of administration	intradermal microdialysis

acetylcholine ()

Name of Sponsor	Anna Stanhewicz
Investigator's Brochure Version	NA
Investigator's Brochure Date	NA
Who is supplying the drug	USP
Who is dispensing the drug	PI/Research Team
Where will the drug be stored	118 PSRB
IND#	124,294
Dose	<0.01
Route of administration	intradermal microdialysis

sodium nitroprusside ()

Name of Sponsor	Anna Stanhewicz
Investigator's Brochure Version	NA
Investigator's Brochure Date	NA
Who is supplying the drug	USP
Who is dispensing the drug	PI/Research Team
Where will the drug be stored	118 PSRB
IND#	124,294
Dose	<0.01mg
Route of administration	intradermal microdialysis

norepinephrine ()

Name of Sponsor	Anna Stanhewicz
Investigator's Brochure Version	NA
Investigator's Brochure Date	NA
Who is supplying the drug	Tocris
Who is dispensing the drug	PI/Research Team
Where will the drug be stored	

IND#	124,294
Dose	<0.01mg
Route of administration	intradermal microdilaysis

angiotensin II ()

Name of Sponsor	Anna Stanhewicz
Investigator's Brochure Version	NA
Investigator's Brochure Date	NA
Who is supplying the drug	Tocris
Who is dispensing the drug	PI/Research Team
Where will the drug be stored	118 PSRB
IND#	124,294
Dose	<0.01mg
Route of administration	intradermal microdialysis

V.9 ***Will any subject be asked to undergo a diagnostic radiation procedure (including radiographic, nuclear medicine, DEXA)?***
No

V.14 ***Will any subject be asked to undergo a radiation therapy procedure (including external beam therapy, brachytherapy, or nuclear medicine therapy)?***
No

V.20 ***Does this project involve the deliberate transfer of recombinant or synthetic nucleic acid molecules, or DNA or RNA derived from recombinant or synthetic nucleic acid molecules, into one or more human research participant?***
No

V.21 ***Will any portion of this project be conducted in the CRU, or does it use any CRU resources?***
Yes

V.22 ***Will this project use:***

- ***any resource/patients of the Holden Comprehensive Cancer Center***
- ***involve treatment, detection, supportive care, or prevention of cancer***

No

V.25.a ***Will the study involve <u>any</u> of the following activity at UI Health Care, even if subjects or their insurance will not be billed for the item or service, and regardless of the study funding source (including studies with departmental or no funding)?***

- ***Procedures, tests, examinations, hospitalizations, use of Pathology services, use of clinic facilities or clinical equipment, or any patient care services, including services conducted in the Clinical Research Unit; or***
- ***Physician services or services provided by non-physicians who are credentialed to bill (ARNPs, Physician Assistants, etc.)***

Yes

V.25.b ***Will there be any procedures or services that may happen as part of a subject's regular medical care and as part of the study?***
No

V.25.c ***Will any study equipment or devices be supplied by a study sponsor?***
No

V.25.e ***Is there or will there be an internal budget for this study?***
No

V.25.f ***Is there or will there be an external budget for this study?***
Yes

V.26 ***The study involves Department of Nursing Services and Patient Care nursing, nursing resources or evaluates nursing practices at UI Health Care.***
No

V.27 ***Will the study involve the use of the I-CTMS (OnCore) for clinical trial data management? Select yes if any or all of the following apply:***

- ***Any study required to register subjects in EPIC are encouraged to use the I-CTMS***
- ***Best practice is to use the I-CTMS for any new study that involves subject tracking or sponsor invoicing***

Note: This question is for non-oncology studies only. For oncology studies use existing HCCC OnCore processes by selecting V.22
No

VI. Subjects

VI.1 ***How many adult subjects do you expect to consent or enroll for this project?***
20

VI.2 ***What is the age of the youngest adult subject?***
18.0

VI.3 ***What is the age of the oldest adult subject?***
50.0

VI.4 ***What is the percentage of adult male subjects?***
0

VI.5 ***What is the percentage of adult female subjects?***
100

VI.6 ***How many minor subjects do you expect to consent or enroll for this project?***
0

VI.13 ***Describe EACH of your subject populations***

- ***Include description of any control group(s)***
- ***Specify the Inclusion/Exclusion criteria for EACH group***

INCLUSION CRITERIA: In this study we are recruiting women who have had preeclampsia in the past 5 years. Post-partum women, 18 years or older, who have delivered within 5 years of the study visit and who have had a preeclamptic pregnancy diagnosed by their obstetrician and confirmed according to the American College of Obstetricians and Gynecologists criteria for preeclampsia. [This information will be self-reported by the subjects.] Because losartan can have dangerous effects on a fetus, women enrolled in this study will be required to use an effective form of birth control [hormonal patch, implant or pill; intrauterine device; consistent use of barrier contraceptive; or prior medical procedure to prevent pregnancy (i.e. tubal ligation, or vasectomy in current partner)].

EXCLUSION CRITERIA: We exclude participants for skin diseases, current tobacco use, diagnosed or suspected hepatic or metabolic disease including chronic kidney disease (CKD) defined as reduced eGFR < 60 mL/min/1.73m2, statin or other cholesterol-lowering medication, current antihypertensive medication, history of hypertension prior to pregnancy, history of gestational diabetes, currently pregnant or planning to become pregnant in the next 6 months, currently breastfeeding, body mass index <18.5 kg/m2, allergy to materials used during the experiment.(e.g. latex), known allergies to study drugs.

VI.14 ***Provide an estimate of the total number of subjects that would be eligible for inclusion in each of your study populations (include your control population if applicable)***
We will recruit subjects from Johnson and surrounding counties in Iowa. According to the Iowa Department of Public Health, Johnson county has a live birth rate of 11.7/1,000 residents or ~1,750 live births per year. Surrounding counties range from 9.6-12 live births/1,000, adding ~4,000 additional live births annually within the geographical area of reach. Assuming an average rate of 7% for preeclampsia incidence, approximately 400 women would be eligible to participate in the preeclamptic group each year. We will recruit women within 5 years postpartum so we estimate that there are approximately 2,000 eligible participants in our geographic area at any given time.

VI.15 ***Describe how you will have access to each of your study populations in sufficient number to meet your recruitment goals.***
We will advertise for participants by posting fliers in the community and in clinicians offices where permission has been granted (see attached flier). We will also post this flier in digital spaces such as on Facebook, Instagram, twitter, and Iowa City Mom's blog (see IC Mom Ad).
Participants will be recruited from the University of Iowa and Iowa City, Iowa by mass email (see attached mass email text) and the Noon News (see attached Noon news posting) a newsletter available on campus at UIHC. We will also recruit participants through Research Match. Lastly, we will recruit participants through the Maternal Fetal Tissue Bank (IRB#200910784). We will obtain the contact information and pregnancy history of the registry participants and reach out to eligible volunteers with the approved recruitment materials (email, flyer, phone call).
All individuals interested in participating will be directed to contact research staff by phone or email. All individuals interested in the study will be invited to complete the RedCap eligibility survey sent via email. If an individual appears to be eligible for the study based on their RedCap Eligibility survey responses, a member of the research team will contact them via phone or email to tell the individual more about the study, answer any questions, and perform a phone screening to determine eligibility.

VI.16 ***Do you <u>plan</u> to recruit/enroll non-English speaking people?***
No

VI.18 ***Do you propose to enroll any of the following in this study as subjects?***

- ***Employee of the PI or employee of a research team member***
- ***Individual supervised by PI or supervised by member of research team***
- ***Individual subordinate to the PI or subordinate to any member of the research team***
- ***Student or trainee under the direction of the PI or under the direction of a member of the research team***

No

VI.20 ***Will subjects provide any information about their relatives?***
No

VI.23 ***Will anyone (other than the subject) provide you with information about the subject (e.g. proxy interviews)?***
No

VI.26 ***Is this project <u>about</u> pregnant women?***
No

VI.27 ***Will this project involve fetuses?***
No

VI.28 ***Does this project involve adult subjects who may be incompetent or have limited decision-making capacity on initial enrollment into the study?***
No

VI.32 ***Does this project involve subjects whose capacity to consent may change over the course of the study?***
No

VI.37 ***Does this project involve prisoners as subjects?***
No

VII.A. Project Description (A)

VII.A.1 ***Where will project procedures take place (check all that apply)?***

- CRU
- Other UI campus site - 118 Pharmaceutical Sciences Research Building (PSRB)

VII.A.2 ***Is this project also being conducted by other researchers at their own sites (e.g. a multi-site collaborative project)?***
No

VII.B. Project Description (B)

VII.B.1. ***Does this project involve any of the following (Check all that apply):***

- ☑ **Interventional** – Includes **Clinical (or Treatment) trial**, **Physiology intervention/study**, **Behavioral intervention/study**, **Diagnostic Trial**.
  - ☑ **Clinical (or Treatment) trial** – A prospective biomedical or behavioral research study of new treatments, new drug or combinations of drugs, new devices, or new approaches to surgery or radiation therapy. (NIH and ClinicalTrials.gov & FDA) The University of Iowa requires a freestanding protocol.
  - ☑ **Physiology intervention/study** – A pharmacologic or measurement study aimed at understanding basic mechanisms of disease and/or of normal human physiology, often without any therapeutic intent (though a clinical trial could include such components, often labeled as "translational" or "basic science" aims.) Measurements in such studies could include, but are not limited to, a blood draw, EKG, EEG, MRI, auditory or sensory testing, checking vital signs, DEXA scans, eye tracking, specimen collection, exercise, fasting, special diets, etc.
  - ☐ **Behavioral intervention/study** – May be used to refer to studies of individual or group behavior. This option does not include drugs, biologics, or devices but could include psychotherapy, lifestyle counseling, behavior modification, etc.
  - ☐ **Diagnostic trial** – Protocol designed to evaluate one or more interventions aimed at identifying a disease or health condition (ClinicalTrials.gov & FDA)
- ☐ **Observational**
- ☐ **Expanded Access** – A process regulated by the Food and Drug Administration (FDA) that allows manufacturers to provide investigational new drugs to patients with serious diseases or conditions who cannot participate in a clinical trial. Examples of expanded access include non-protocol access to experimental treatments, including protocol exception, single-patient IND, treatment IND, compassionate use, emergency use, continued access to investigational drug, and parallel track (ClinicalTrials.gov & FDA).
- ☐ **Registry** – The collection and maintenance of data (not including biologic samples) in which: (1) the individuals in the registry have a common or related condition(s), and/or (2) the individuals in the registry are interested in being contacted for future studies by investigators other than those listed in Section II of this project.(UI Guide)
- ☑ **Repository** – The collection, storage, and distribution of human biologic samples and/or data materials for research purposes. Repository activities involve three components: (i) the collection of data and/or specimens such as blood, tissue, saliva, etc.; (ii) the storage of data or specimens, and data management function; and (iii) the sharing of data/specimens with recipient investigators other than the original investigators. (paraphrased from OHRP)
- ☐ **Other**

VII.B.1.a ***Does this project involve any of the following (Check all that apply):***

- ☐ **Phase I trials** – include initial studies to determine the metabolism and pharmacologic actions of drugs in humans, the side effects associated with increasing doses, and to gain early evidence of effectiveness; may include healthy participants and/or patients (ClinicalTrials.gov & FDA)
- ☐ **Phase II trials** – include controlled clinical studies conducted to evaluate the effectiveness of the drug for a particular indication or indications in patients with the disease or condition under study and to determine the common short-term side effects and risks(ClinicalTrials.gov & FDA)
- ☐ **Phase III trials** – include expanded controlled and uncontrolled trials after preliminary evidence suggesting effectiveness of the drug has been obtained, and are intended to gather additional information to evaluate the overall benefit-risk relationship of the drug and provide an adequate basis for physician labeling(ClinicalTrials.gov & FDA)

- ☐ **Phase IV trials** – studies of FDA-approved drugs to delineate additional information including the drug's risks, benefits, and optimal use(ClinicalTrials.gov & FDA)

VII.B.1.c ***What is the last date the final subject/participant will undergo an intervention (i.e. the last study visit by any subject) as required by the protocol?***
08/31/2024

VII.B.2 ***Does this project involve a drug washout (asking subject to stop taking any drugs s/he is currently taking)?***
No

VII.B.6 ***Will any subjects receive a placebo in this study when, if they were not participating, they could be receiving an FDA-approved treatment for their condition?***
No

VII.B.11 ***Is there a separate, written protocol that will be submitted in addition to this IRB New Project form? (Note: a grant application is not considered to be a protocol)***
No

VII.B.18 ***Does this project involve testing the safety and/or efficacy of a medical device?***
No

VII.C. Project Description (C)

VII.C.1 ***Does this project involve any research on genes or genetic testing/research?***
No

VII.D. Project Description (D)

VII.D.1 ***Check all materials/methods that will be used in recruiting subjects (you will need to attach copies of all materials at the end of the application):***

- Website - https://microvascularphys.lab.uiowa.edu/research-studies https://clinicaltrials.uihealthcare.org/studies/angiotensin-ii-receptor-inhibition-improve-microvascular-function-women-who-have-had https://iowacity.momcollective.com/ https://www.facebook.com/iowacitymoms/ https://www.instagram.com/iowacitymoms/ https://twitter.com/AnnaStanhewicz?t=x574TViYU-Nuc9ryBH8Mdw&s=09
- Letter -
- Advertisements -
- Posters -
- E-mail -
- Existing Registry/database - We will recruit participants through the Maternal Fetal Tissue Bank (IRB#200910784). We will obtain the contact information and pregnancy history of the registry participants and reach out to eligible volunteers with the approved recruitment materials (email, flyer, phone call).
- Research Match.org -

VII.D.1.a ***Will any of the materials/methods below be used by researchers (or their colleagues) to recruit subjects into this study?***

- ***the potential subject is a patient OR***
- ***use of any information considered to be Protected Health Information (PHI) OR***
- ***review of patient/clinic records be used in recruiting subjects***

Yes

VII.D.1.b ***Describe source of records***
Patient EPIC records will be reviewed and screened for suitability. Potential subjects name, contact information, age, and pregnancy history will be reviewed. This will be accomplished by starting with a TriNetX exchange utilizing ICD9 and 10 codes and inclusion criteria to obtain MRN.

VII.D.1.c ***Select all Private Identifiable Information (PII) or Protected Health Information (PHI) accessed and used for this study (select all that apply)***

Identify types of PHI accessed

Type of PHI	Data source
Name	☑
Street address	☑
City	☑
County	☐
Precinct	☐
Zip code	☑
Geocodes smaller than state	☐
Date of birth, ages > 89 years of age	☐
Diagnosis dates	☐
Procedure dates	☐
Admission or discharge dates	☐
Telephone numbers	☑

Type of PHI	Data source
Fax numbers	☐
E-mail addresses	☐
Social Security number	☐
Medical record number	☑
Health plan beneficiary or account numbers	☐
Certificate/license numbers	☐
Vehicle identifiers and serial numbers or license numbers	☐
Device identifiers or serial numbers	☐
Web URLs	☐
Internet Protocol (IP) address numbers	☐
Biometric identifiers including finger/voice prints	☐
Full face photographic images or any comparable images	☐
None of the above	☐

VII.D.2.a ***List ALL of the variables, including any identifiers not previously entered or links to identifiers you plan to obtain/use for purposes of this study. (The information accessed should be the minimum data variables necessary for performing the desired analysis.)***
Name, age, pregnancy history (including preeclampsia diagnoses in the past 5 years), pregnancy status, and current medication use. We will also need to look at data elements that correspond with our exclusion criteria including skin diseases, current tobacco use, diagnosed or suspected hepatic or metabolic disease including chronic kidney disease (CKD) defined as reduced eGFR < 60 mL/min/1.73m2, statin or other cholesterol-lowering medication, current antihypertensive medication, history of hypertension prior to pregnancy, history of gestational diabetes, currently pregnant or planning to become pregnant in the next 6 months, currently breastfeeding, body mass index <18.5 kg/m2, allergy to materials used during the experiment.(e.g. latex), known allergies to study drugs.

VII.D.3 ***Describe why you could not practicably recruit subjects without access to and use of the information described above***
The study requires the subject to meet very specific inclusion/exclusion criteria on their diagnosis. It is easier on the subject to determine this by reviewing their medical records. It will save the subject the time and effort coming to the study site just to learn the do not qualify based on current information.

VII.D.4 ***Describe why you could not practicably obtain authorization from potential subjects to review their patient or clinic records for recruitment purposes.***
Without reviewing the medical record information pertaining to inclusion/exclusion criteria we would not be able to identify subjects who may qualify. By reviewing the potential subject's medical record and identifying possible inclusion/exclusion criteria, the study team will be able to determine if a subject has a possibility of qualifying which may save the subject and study team the time and effort to participate in a visit that may result in the subject not qualifying. Also, it would not be practicable to approach all individuals presented at the clinic to ask if their medical record could be reviewed to determine eligibility for the research.

VII.D.5 ***Describe plans to protect the identifiers from improper use or disclosure***
We will only collect the minimum amount of information needed to determine eligibility. We will review the information visually first. All information will be kept in a study specific folder only accessible to study staff. Once the subject qualifies, all data is collected under the study specific ID. Names and the pre-screening forms of subjects that do not qualify or decline will be kept until enrollment has ended, so that the study team does not approach them again.

VII.D.6 ***Describe plans to destroy identifiers at the earliest opportunity consistent with conduct of the research***
Once consent is signed, the subject will be assigned a study ID. From that point on, only the study ID will be used. Once enrollment has ceased, the names and pre-screening forms of potential subjects that did not enroll in the study will be destroyed by secure shred.

VII.D.7 ***Does the research team agree that the requested information will not be reused or disclosed to any other person or entity, except as required by law, for authorized oversight of the study, or for other research for which the use or disclosure of the requested information would be permitted by the HIPAA Privacy Rule***
Yes

VII.D.8 ***Will a member of the research team discuss the study with the subject in person prior to the subject agreeing to participate?***
Yes

VII.D.9 ***Describe the physical location where the consent process will take place:***
The research staff will discuss the study with potential subjects in a conference room or exam room in the ICTS clinical research unit or in the research lab (118 PSRB).

VII.D.10 ***Will a member of the research team discuss the study with the subject by phone prior to the subject agreeing to participate?***
Yes

VII.D.11 ***Describe:***
Research staff will discuss the project over the phone with individuals who are interested in participating in the study. (see pre-consent phone script). This will take place in the research office in 118 PSRB. During the call, the research team member will answer any questions and describe the study to the individual as needed. If the subject is still interested in participating they will be given access to the RedCap pre-consent screening survey. If the individual continues to meet eligibility after completing this survey, they will be given information about the study, but will not be consented over the phone. If the subject agrees, they will come to either the clinical research unit or 118 PSRB and be consented and proceed with Visit 1.

Also, the study team will use TriNetX exchange utilizing ICD9 and 10 codes and inclusion criteria to obtain MRNs of individuals who appear to meet eligibility criteria. These potential subjects will be sent a letter (see recruitment letter) briefly explaining the study and asking the potential subject to call the study team. The letter will also state that a phone call will be attempted up to three times to the potential subject two weeks following the letter being sent if the study team does not hear from the potential subject. In that phone call, the subject will be asked if they are interested in hearing about a study they may qualify for. If they are interested, the study team will explain the study in detail, including all study procedures and time commitments (see pre-consent phone script). If the potential subject decides they want to proceed

they will be asked questions from the pre-screening form and if subjects appears they might qualify, an appointment will be scheduled for consent and screening visit as described in the above paragraph.

VII.D.12 ***Who will be involved in the consent process (including review of consent document, answering subjects' questions)?***

Name	Consent Process Involvement
Anna Reid-Stanhewicz, PHD	Yes
Adam Corkery, PHD	Yes
Vincenzo Dimarco, High School	No
Alyssa Engels, High School	No
Ashley Faber, High School	No
Katharine Geasland, BSN	No
Claire Goebel, BA	Yes
Diana Jalal, MD	No
Isabel Klimowicz, High School	No
Ruda Lee, MS	Yes
Maddie Marino, High School	No
Grace Maurer, MS	Yes
Leah Patton, High School	No
Gary Pierce, PHD, MS	No
Bomi Ryang, BS	No
Kelsey Schwartz, BS	Yes
Nikolas Soofi, High School	No
Amy Marie Stroud, MSN	No
Olivia Taeger, High School	No
Lizzy Wetzel, High School	No
Ellie Wilson, High School	No

VII.D.15 ***Check all materials that will be used to obtain/document informed consent:***

- Consent Document
- Consent Summary (or Key Information Sheet)

VII.D.16 ***Are you requesting a waiver of documentation of consent (either no subject signature or no written document)?***
No

VII.D.19 ***Before the subject gives consent to participate are there any screening questions that you need to directly ask the potential subject to determine eligibility for the study?***
Yes

VII.D.20 ***List any screening questions you will directly ask the potential subject to determine eligibility.***
Eligibility Survey: (see attached pre-consent online survey) This survey will be administered via REDCap link delivered by email. Participants will be invited to complete a survey to determine their eligibility.

If subjects' responses to the online screening survey deem them eligible, they will be contacted by a member of the research team via phone to confirm their answers to the pre-consent online survey, answer additional questions, and schedule visit 1.

VII.D.21 ***Will you keep a screening log or other record that would include information on people who do not enroll in the study?***
Yes

VII.D.22 ***Describe the information being collected and the purpose for keeping this information.***
The following information will be collected in a screening log:
1. Subject's name
2. Age
3. How they heard about study
4. Date that they contacted the study
5. How they contacted us (phone/email)
6. Date of phone screening
7. Phone number
8. Email address
9. Pass online screening; yes or no
10. If did not pass online screening, reason?
11. Pass phone screen? Yes or No
12. If did not pass phone screen, reason?
13. If passed phone screening, date of consent
14. Signed informed consent

Contact information is required to contact the subjects after the phone screening in case the research staff needs to reschedule Visit 1 or for follow up if the subject does not show for the Visit 1. Because this study is recruiting using mass email, subject information including a brief description why they were deemed ineligible (i.e. health history, medication, breastfeeding) will be kept to prevent making contact twice with an individual who is ineligible. Information on why a potential subject is ineligible/not passing the phone screening is to report to NIH and to monitor our recruiting progress. Information on how the subjects heard of the study will help the research team understand the most successful methods for advertising for the study.

VII.D.23 ***Will this information be shared with anyone outside the UI research team members?***
No

VII.D.25 ***After the subject agrees to participate (signs consent), are there any screening procedures, tests, or studies that need to be done to determine if the subject is eligible to continue participating?***
Yes

VII.D.26 ***List and describe screening***
Complete the additional info survey (see attached RedCap document "additional info survey") for study records
Medical history and physical exam
Resting blood pressure and heart rate
Standard blood chemistries (lipids, complete blood count, basic metabolic panel, liver enzymes)
Urine pregnancy test

VII.D.27 ***Discuss how much time a potential subject will have to agree to consider participation and whether or not they will be able to discuss the study with family/friends before deciding on participation.***
There is no time limit for the subject to agree to consider to be in the study as long as the study is actively recruiting subjects and they are still eligible. Subjects are allowed to discuss the study with family/friends before deciding on participation.

VII.D.28 ***How long after the subject agrees to participate do study procedures begin?***
The procedures in visit 1 (screening visit) can occur on the same day as consent. The experimental visits (pickup and begin oral treatment) will begin within 1-2 weeks or less of visit 1.

VII.D.29 ***Provide a description of the enrollment and consent process for adult subjects***

- ***Describe each study population separately including control population***
- ***Include when recruitment and consent materials are used***
- ***Use 3rd person active voice "The Principal Investigator will identify subjects. For example, the principal investigator will identify potential subjects, the study coordinator will discuss the study with subjects over the telephone and schedule the first study visit, etc..."***
- ***Describe the steps that will be taken by the research team to minimize the possibility of coercion or undue influence during the consent process***

The subjects will consist of healthy women (age 18 and older) who have delivered a baby within the past 5 years and had preeclampsia during their pregnancy, but were free of cardiovascular or metabolic disease prior to pregnancy. These subjects will be enrolled and complete losartan and placebo treatments in a double-blind randomized crossover design. The PI and research staff will recruit subjects from Johnson county and surrounding areas via advertisements, flyers, emails, The UI mass-email system, newspaper and newswire ads, Research Match.org, obtaining contact information from the Maternal Fetal Tissue Bank (MFTB) registry, utilizing TriNetX (discussed in more depth below), and posting on the UIHC website for research volunteers. Recruitment materials (attached MFTB recruitment letter) will be sent to women registered with the MFTB who fit our inclusion criteria and have consented to be contacted for future studies. The UIHC Clinical Trials and Research Website will have this research study available as a listed research study for potential research subjects to find out more information and express interest in participating. Additionally, paid advertisements (see attached IC Mom Ad) will be posted on the Iowa City Moms website and social medias. These advertisements when clicked will link to the study's page on the UIHC Clinical Trials and Research website.

All potential subjects will be asked to contact the research staff via phone or email. A study team member will then contact the potential participant by phone to complete the phone screening and schedule visit 1 if they are eligible and wish to participate. Subjects will be sent the consent summary either by e-mail or postal mail (subject preference) before visit 1. Visit 1 will include a detailed description of the study and review of the informed consent, including risks. If subjects are not able to understand the protocol and instructions for any reason, written or verbal, they are not included in the study. Subjects are informed throughout the consenting, screening, and conduction of the study that they may discontinue their participation at any time with no penalty to them. If the subject signs the informed consent, they are given a copy of the signed document to take home with them.

Research staff will discuss the project over the phone with individuals who are interested in participating in the study. (see pre-consent phone script). This will take place in the research office in 118 PSRB. During the call, the research team member will answer any questions and describe the study to the individual as needed. If the subject is still interested in participating they will be given access to the RedCap pre-consent screening survey. If the individual continues to meet eligibility after completing this survey, they will be given information about the study, but will not be consented over the phone. If the subject agrees, they will come to either the clinical research unit or 118 PSRB and be consented and proceed with Visit 1.

The study team will use TriNetX exchange utilizing ICD9 and 10 codes and inclusion criteria to obtain MRNs of individuals who appear to meet eligibility criteria. These potential subjects will be sent a letter (see recruitment letter) briefly explaining the study and asking the potential subject to call the study team. The letter will also state that a phone call will be attempted up to three times to the potential subject two weeks following the letter being sent if the study team does not hear from the potential subject. In that phone call, the subject will be asked if they are interested in hearing about a study they may qualify for. If they are interested, the study team will explain the study in detail, including all study procedures and time commitments (see pre-consent phone script). If the potential subject decides they want to proceed they will be asked questions from the pre-screening form and if subjects appears they might qualify, an appointment will be scheduled for consent and screening visit as described in the above paragraph.

VII.D.37 ***Does the study include any form of deception (e.g., providing participants with false information, misleading information, or withholding information about certain study procedures)?***

Examples:

- ***Procedure includes a cover story that provides a plausible but inaccurate account of the purposes of the research.***
- ***Participants will be provided with false information regarding the particular behaviors of interest in the research.***
- ***Procedures include a confederate pretending to be another participant in the study.***
- ***Participants will be told that the research includes completion of a particular task, when in fact, that task will not be administered.***
- ***Study is designed to introduce a new procedure (or task) that participants are not initially told about.***
- ***If yes, a waiver of informed consent must be requested under question IV.3.***

No

VII.E. Project Description (E)

VII.E.1 ***Will subjects be randomized?***
Yes

VII.E.1.a ***Will any subjects be blinded to which study arm they have been assigned?***
Yes

VII.E.1.b ***Does the protocol permit telling subjects their treatment assignment at the end of the entire study?***
Yes

VII.E.1.c ***Describe the circumstances under which subjects will be told what study arm they have been assigned.***
After the study is completed, subjects can be told which treatment they received at which visit if they wish to know.

VII.E.2 ***Describe randomization scheme/assignment including ratio such as 1:1, 2:1 etc.***
Subjects will be randomized to which treatment they receive first (placebo or losartan). All subjects will complete both arms of the study. They will be blinded to which treatment they are receiving at the time of the treatments. If they wish to know, subjects can be informed which treatment they received at what time when they complete the study.

VII.E.3 ***Will any questionnaires, surveys, or written assessments be used to obtain data directly from subjects in this study?***
Yes

VII.E.4 ***List all questionnaires, surveys, written assessments and ATTACH each one to the application. (NOTE: You are NOT prohibited from attaching copyrighted materials to this application)***
pre-consent screening survey
additional information survey
health history questionnaire

VII.E.5 ***Does this project involve creating any audiotapes, videotapes, or photographs?***
No

VII.E.6 ***Provide a detailed description in sequential order of the study procedures following the consent process - DO NOT cut and paste from the Consent Document.***

Describe study populations separately if they will be participating in different procedures - include CONTROL population if applicable.

DESCRIBE:

- ***What subjects will be asked to do/what happens in the study (in sequential order)***
- ***The time period over which procedures will occur***
- ***The time commitment for the subject for individual visits/procedures***
- ***Long-term followup and how it occurs***

Visit 1 - Consent and Screening (approximately 1 hour)
The PI or research staff explain the study and go over the informed consent with the participant. After the participant signs the informed consent, the PI or research staff performs the physical screening that includes urine pregnancy test, heart rate (HR), blood pressure (BP), height, waist circumference, and weight measurements. The participant fills out the health history questionnaire and confirms that they are using an effective form of birth control. A research nurse performs or approved laboratory personnel standard venipuncture to obtain blood (total = 7.5ml blood) for complete blood count (CBC), chemistry analysis, and lipid profile to be measured at UIHC pathology labs. The subject provides a urine sample for the measurement of urine proteins which will be measured by the UI Diagnostic Lab.

Visits 2 and 5- Pick up study medications (approximately 15 minutes)
The PI or research staff give the participant study medication (losartan or placebo). The PI or research staff goes over the instructions for taking the study medication which includes how many pills to take and at what times of day, potential side effects, and who to call if they have questions or problems while taking the medication.

Visits 3 and 6 - Pick up 24 hour blood pressure monitor (approximately 15 minutes)
The PI or research staff instrument the participant with the 24 hour blood pressure monitor. The PI or research staff goes over the instructions for wearing the monitor.

Visits 4 and 7 - Experimental visits (approximately 4 hours each)
Note: prior to coming to PSRB for the experimental visits, subjects take the oral study medication for 6 weeks. Subjects will ingest 1 pill per day. After the first study visit, subjects will undergo a washout period of at least 14 days in which they do not ingest any study medication, before beginning the same 6 week regimen of the other treatment.

In the event of an experimental failure (e.g. equipment stops working, building power outage during study visit, etc) subjects may be asked to repeat a trial.

Experimental procedures
24 hours before the study visit, subject will be instrumented with a 24 hour blood pressure monitor to wear home for 24 hours. Subject will return the blood pressure monitor to the investigators when they come in for the experimental visit.

Subjects will come to the CRU or 118 PSRB where a research nurse or approved laboratory personnel performs a blood draw (14.5 ml, < 1 Tablespoon) for substances of interest (e.g. inflammatory cytokines) and immune cell activity, and blood chemistry. The subject provides a urine sample for the measurement of urine proteins which will be measured by the UI Diagnostic Lab. Then the rest of the study visit occurs in 118 PSRB for the experiment (Pulse wave velocity and intradermal microdialysis).

Pulse wave velocities (PWV) on Carotid-femoral and carotid-brachial: Carotid-femoral artery PWV, the ‘gold standard’ in vivo assessment of aortic

stiffness in humans, will be assessed by applanation tonometry using the Sphygmocor pulse wave analysis system (AtCor Medical, Inc.) as previously described by the PI.45-47 After 10 minutes of supine rest, carotid, brachial, and femoral artery pulse waveforms will be recorded non-invasively by sequentially recording of carotid, brachial, and femoral artery pulse waveforms with a hand-held transducer for 20 seconds. Pressure waveforms are gated to the 13 lead ECG R wave in order to calculate the transit time (t) between the foot of the carotid and the femoral waveforms.85, 86 The carotid-femoral and carotid-brachial transit distance (CFTD) is estimated between the 2 anatomical sites as the difference between the suprasternal notch (SSN) to carotid (SSN-C) and femoral (SSN-F) sites. Thus, the CFTD=(SSN-F)-(SSN-C) and PWV calculated as CFTD/t. This approach accounts for parallel transmission of the pulse wave up the brachiocephalic and carotid arteries, and simultaneously along the aortic arch using the SSN as a fiducial point where parallel transmission begins (e.g., bifurcation site of aortic arch/brachiocephalic artery).

Microdialysis Probe Insertion: (NOTE: The intradermal microdialysis procedure used here is identical to the one approved in IRB #201909818) Please see the attached "intradermal microdialysis" document for general information about intradermal microdialysis, schematic representation of the procedure, and references. The researchers place a tight band around the forearm so they can visualize veins. For each MD site, they make pairs of pen-marks on the arm 2.5 cm (1 inch) apart and away from veins. They remove the tight band. The MD tubing enters and exits the skin at the marks. The researchers clean the arm with povidone iodine and alcohol, and place an ice bag on the site for 5 minutes to numb the skin. Then they insert a thin needle into the skin at each entry mark. The needle's tip travels between the layers of skin for 2.5 cm (1 inch) and exits the skin at the matching exit-mark. They thread the microdialysis tubing through the needle and then withdraw the needle leaving the tubing in the skin. Any hyperemia related to the insertion subsides in about 60 minutes. When the hyperemia induced by inserting the tubing in the skin subsides, the experiment begins. During this time Lactated Ringer's perfuses the tubing. The researchers tape a fiber optic laser Doppler flowmeter probe and its holder over each microdialysis site. The researchers control the temperature of the holders. The holders start at 33°C (91.4°F). During the experiment, the computerized data acquisition system records heart rate, skin blood flow, and skin temperatures continuously. The researcher and/or an automated critical care monitor measures blood pressure at 5-7 minute intervals.

Acetylcholine, Angiotensin II, and Norepinephrine Dose Responses: The intradermal microdialysis probes are randomly numbered 1 - 4 and assigned
to receive doses of acetylcholine with or without L-NAME (probes 1 and 2), angiotensin II (probe 3) or norepinephrine (probe 4).
Acetylcholine Dose Response: Microdialysis Probe 1. Lactated Ringer's only
Microdialysis Probe 2. Lactated Ringer's + L-NAME
Angiotensin II Dose Response: Microdialysis Probe 3. Lactated Ringer's only
Norepinephrine Dose Response: Microdialysis Probe 4. Lactated Ringer's only

When the skin blood flow is stable, the researchers perform a 10-minute baseline set of measurements. The researchers add identical concentrations of acetylcholine to the perfusate in probes 1 and 2 and Angiotensin II to the perfusate in probe 3 and Norepinephrine to the perfusate in probe 4. At 5-minute intervals, they increase the concentration of acetylcholine, angiotensin II or norepinephrine in the perfusates. The participant receives 10 different concentrations of acetylcholine and 9 different concentrations of angiotensin II or norepinephrine. After perfusing the last concentrations, the researchers warm the temperature controllers to 43°C (108°F) and switch perfusates at all sites to lactated Ringer's for about 30 minutes. After 30 minutes they add SNP to the perfusates for approximately 5 minutes. Heating and SNP perfusion causes maximum vasodilation at the microdialysis site. The researchers then de-instrument the subject, remove the microdialysis fibers, and apply
sterile dressings. The subject is given verbal and written instructions for how to care for the microdialysis sites. The researcher measures final vitals before the subject departs. At the end of visit 2, the subject is given instructions for the 14 day washout period. The subject may be given the pills for the second treatment at the end of visit 2(replacing visit 4), along with instructions for when and how to take the pills prior to study visit 5.

VII.E.7 ***Will you attempt to recontact subjects who are lost to follow-up?***
No - followup is not required in this study

VII.E.9 ***Will subjects be provided any compensation for participating in this study?***
Yes

VII.E.10 ***Cash***
No

VII.E.11 ***Gift Card***
No

VII.E.12 ***Check***
Yes

VII.E.13 ***Who will be providing the research compensation check to the subject?***
Accounting Services directly via the e-Voucher system

VII.E.16 ***Other***
Yes

VII.E.17 ***Describe:***
Parking vouchers will be provided during study visits.

VII.E.18 ***If you plan to compensate subjects using cash, checks or cash equivalent does your unit have a Cash Handling Procedure in place that has been approved by Accounting Services?***
Yes

VII.E.19 ***Describe the compensation plan including***

- ***Compensation amount and type per visit***
- ***Total compensation***
- ***Pro-rating for early withdrawal from study***

Pre-treatment: $30 per treatment ($5/week for 6 weeks)
24 hour blood pressure monitor: $10
Experimental visits: $100 per visit ($15/microdialysis probe + $10 for pulse wave velocity measurements + $7.50/hour for completing the experiment)

Total for completing the study: $280 (two pre-treatment periods, two 24 hour blood pressure monitors, two completed study visits)
Parking pass for time involved at study and screening visits.

Pro-rating: Subjects can receive payment for pre-treatment and experiments not completed. The researchers pay an amount of money equal to the part completed. For instance, if a subject completes half of Experiment 1, the subject receives $15.00 for each probe inserted plus $7.50 for each hour they completed. The researchers may ask subjects to repeat a trial. If subjects agree to repeat a trial, they receive payment for the repeated trial as stated above.

VIII. Risks

VIII.1 ***What are the risks to subjects including***
- emotional or psychological
- financial
- legal or social
- physical?

Physical Risks:
Microdialysis: Intradermal microdialysis is a specialized research technique. Dr. Stanhewicz is highly skilled in this technique and has been using the procedures in this protocol (including fiber placement, perfusate preparation, subject monitoring, and fiber removal) for over 10 years with no adverse events and have been reviewed and approved by Dr. Diana Jalal at the University of Iowa.

Cutaneous microdialysis commonly causes some pain and bruising similar to that experienced during venipuncture. There is usually no pain after the probe is in place. The participant may experience mild pain while the researchers remove probe. Minor bleeding may occur. As with any event that breaks the skin, infection is possible. However, no participants in any of the researchers' prior experiments have reported infection. In the unlikely event in which the membrane breaks during removal leaving an isolated piece of membrane under the skin, they treat the piece of membrane in a manner similar to that for a splinter in the skin. In this case, trained personnel make a superficial incision for removal. Such an event has never occurred in any projects overseen by Dr. Stanhewicz.

Microdialysis delivers small amounts of pharmacological substances to a nickel-sized area of the skin. Therefore there are no individual risks associated with the drugs perfused through the fibers (acetylcholine, L-NAME, angiotensin II, norepinephrine, SNP, lactated Ringer's). The small quantities used and the extremely localized administration during microdialysis does not produce systemic effects. To the researchers' knowledge, there are no reports of long or short-term side effects of these substances administered through microdialysis. The chance of adverse reactions to these substances is extremely small given the minute amount delivered to the a very small area of skin, the lack of adverse reactions to similar amounts delivered via MD in many other studies, and lack of adverse effects in human cell cultures. There is a slight chance of allergic reaction to these substances that could produce redness, itching, rash, and/or swelling.

Local Heating: The local heating control unit (Moor Instruments) precisely controls and monitors the temperature of the heated probe holders used with the Laser Doppler Flowmeter. The system has programmed maximum temperature limits. To determine the maximal SkBF, the researchers increase the temperature of the heating units slowly (about 0.1°C every 1 second). The skin feels very warm but not painful. Local heating causes temporary redness of the skin that subsides within several hours. This technique is very unlikely to produce long-term ill effects.

Povidone Iodine: Hospitals and researchers use povidone iodine to clean and sterilize the skin. Participants could be allergic to iodine. An allergic reaction could cause redness, itching, rash, and/or swelling. Staff use only alcohol on participants with iodine allergy as identified during screening.

Tape and adhesive disks: Participants could be sensitive to the adhesive of the tape, ECG electrodes, and double-sided adhesive disks used in the study causing redness, rash, tenderness, and/or itching. The researchers remove these items carefully.

Blood draws: Potential risks associated with obtaining blood samples are minimal but include slight bruising, pain, a temporary feeling of faintness, and/or a small risk of infection. All blood draws will be performed by a research nurse or team member trained in drawing blood.

24-hour blood pressure measurement: There are no known or foreseeable risks associated with 24-hour blood pressure monitoring. The hourly cuff inflation may be disruptive to the participants activities of daily life. The cuff may cause mild pressure discomfort when inflating.

Pulse wave velocity (PWV): There are no known or foreseeable risks associated with the use of applanation tonometry for PWV. EKG patches are used for the monitoring and may cause minor skin irritation.

Losartan Pharmacotherapy: Losartan is a commonly prescribed hypertensive medication that works by blocking the angiotensin II type 1 receptors located on the vascular tissues. The most common but least serious side effects of losartan are cold symptoms (stuffy/runny nose, sneezing, sore throat), mild dizziness, back pain, and diarrhea.

Injury or death of unborn babies: Losartan is classified as pregnancy category D. Studies in pregnant women have demonstrated risk to the fetus. Pregnancy or planning to become pregnant are exclusion criteria in this study.

Low blood pressure (hypotension): Use of an anti-hypertensive medication may cause a decrease in blood pressure that leads to dizziness, faintness, or lightheadedness, especially when going from lying down to standing up. This can be treated by stopping the medication.

For people who already have kidney problems, losartan may lead to worsening of kidney function. Abnormal kidney function is an exclusion criteria for this study. We screen participants for markers of kidney function, and repeat those measurements (blood chemistry) at the end of each treatment period.

Swelling in feet, ankles, or hands, or unexplained weight gain: In some patients, losartan use may lead to edema. This can be treated by stopping the medication.

Allergic reaction: Subjects could have a mild or severe allergic reaction including hives; difficult breathing; swelling of the face, lips, tongue, or throat. We exclude subjects who have a known allergy to losartan.

Emotional or Psychological risks: There are no foreseeable psychological risks with this study.

Social Risks: There are no foreseeable social risks with this study

Legal Risks: There are no foreseeable legal risks with this study.

Loss of confidentiality is possible.

VIII.2 ***What have you done to minimize the risks?***

- ***If applicable to this study ALSO include:***
  - ***How you (members of your research team at Iowa) will monitor the safety of individual subjects.***
  - ***Include a description of the availability of medical or psychological resources that subjects might require as a consequence of participating in this research and how referral will occur if necessary (e.g. availability of emergency medical care, psychological counseling, etc.)***

Note: All investigational substances used in these protocols are used with general Physician Oversight (see attached Standard Operating Procedures document)

Risks Associated with Losartan Treatment: Because of the risk of injury or death to unborn babies we perform a pregnancy test on all subjects before administering losartan and council participants on the risks of becoming pregnant while taking the medication. During the study if a subject experiences side effects that are life threatening, the subject will be instructed to call 911. If the subject experiences any mild but tolerable expected side effects such as cold symptoms, mild dizziness, back pain, or diarrhea, they will be asked to keep a log of these including the date, the duration, and the severity by rating on a scale of 1 (mild) to 10 (severe/intolerable). If the subject feels the side effects are uncomfortable or intolerable, then they will be instructed to call the Research Associate (Claire Goebel) or Dr. Stanhewicz. The Research Assistant or Dr. Stanhewicz (although she will be blinded) will instruct the subject to skip the next dose and if the symptoms/side effects have resolved at time of next dose (in 24 hours), they will be instructed to take the next dose. Dr. Stanhewicz will consult with Dr. Jalal if necessary. If symptoms do not resolve or worsen at time of next scheduled dose, the subject will be instructed to discontinue the treatment. The subject will be withdrawn from the study procedures but will be followed up as necessary.

Risks Associated with Intradermal Microdialysis: Research techniques are only performed by personnel who are trained and approved by Dr. Stanhewicz to complete these procedures. Dr. Stanhewicz has 10 years of experience utilizing intradermal microdialysis without unanticipated adverse events. All laboratory personnel are trained in CPR and basic first aid. In the event of a life threatening emergency, lab personnel call 911. The Field House is equipped with AED and lab personnel are trained in how to use them if necessary. Prior to placing the microdialysis fibers, the researcher puts on sterile gloves and cleans the skin with iodine and alcohol to reduce the risk of infection. The researchers apply ice to the skin for 5 minutes before placing the microdialysis fibers to reduce any pain associated with placement. Once the fibers are in place, research personnel constantly monitor subjects for adverse reactions (e.g. pain, itching, redness, swelling) to the microdialysis fibers or the perfusates. Participants are never left unsupervised with the fibers in place. An automated monitor measures blood pressure every 5 minutes and constantly records heart rate. The researchers clip the ends of the fibers and clean them with an alcohol swab before removing them from the skin to reduce the risk of infection. They stop any bleeding by applying mild pressure to the sites with a sterile gauze pad. The researchers apply a clean, sterile bandage to the area before the participant leaves. Participants are given verbal and written instructions on how to care for the sites and to call Dr. Stanhewicz or the research nurse if they have any questions or concerns about the sites after they have left the study visit.

Risk of loss of confidentiality: The investigators collect the minimum amount of confidential data in order to complete the aims. All subjects are assigned a code and data and specimens are collected using the code only. The only time subject names and codes appear together is on their file which is kept in a locked cabinet in the PI's office. Only IRB approved personnel have access to these files. All data files are stored on password protected computers and servers and only IRB approved personnel have access. Extensive details on how the risk of loss of confidentiality is minimized are provided in section X. Privacy and Confidentiality.

VIII.3 ***Does this study have a plan to have an individual or committee review combined data from all subjects on a periodic basis (such as summary or aggregate safety and/or efficacy data)?***
Yes

VIII.4 ***Describe the plan to review combined data from all subjects, such as summary or aggregate safety and/or efficacy data. Include the following:***

- ***Describe what data will be summarized and reviewed***
- ***Describe how frequently data will be reviewed.***

The local Data Safety Monitoring Board performs bi-annual reviews of the study protocol, subject enrollment information, and aggregate data collected to that point. The DSMB does not have access to individual identification of subjects. The DSMB then makes recommendations regarding the progress of the study.

VIII.5 ***Will overall safety monitoring be performed by individual(s)/committee at The University of Iowa. (NOTE: If this study involves more than minimal risk, in most cases these should be individuals who are not members of the study research team.)?***
Yes

VIII.6 ***List names:***
Mark Santillan, MD; Melissa Swee, MD; Bridget Zimmerman, PhD

VIII.7 ***Will overall safety monitoring be performed by individuals or committee not associated with The University of Iowa (such as a study Data Safety Monitoring Board)?***
No

IX. Benefits

IX.1 ***What are the direct benefits to the subject (do not include compensation or hypothesized results)?***
The study procedures and/or findings do not provide direct benefits to the subjects participating in the study. Subjects receive a medical screening that could inform them about their health. They learn their blood pressure and blood cholesterol levels. This is important knowledge. High blood pressure and blood cholesterol contribute to many serious health problems. We advise those with high blood pressure or blood cholesterol to follow-up with a health care provider. They also learn of the connection between
preeclampsia and cardiovascular disease.

IX.2 ***What are the potential benefits to society in terms of knowledge to be gained as a result of this project?***
Preeclampsia, a disorder of pregnancy effecting ~5-8% of pregnancies in the United States, and ~8 million pregnancies worldwide. Otherwise healthy women who develop preeclampsia during pregnancy have 2-4 times greater risk for developing cardiovascular disease later in life. Cardiovascular disease is the leading cause of death among women worldwide, a fact lending even more gravity to the need for elucidating the mechanisms mediating the vessel dysfunction associated with preeclampsia and the increased risk for cardiovascular disease. The information to be gained is essential to the understanding of the related cardiovascular pathology. This knowledge could suggest and aid the development of novel therapeutic strategies for the management of risk in this population.

X. Privacy & Confidentiality

X.1 ***What are you doing to protect the privacy interests of the subjects?***
The minimum amount of data necessary to complete the aims will be collected during the study. The informed consent process will be conducted in a private exam room in the CRU or the research office in 118 PSRB with the door closed. All screening and experimental procedures will be conducted in the laboratory in 118 PSRB with the door closed and locked. Only personnel directly involved in the study will be allowed in the rooms.

X.2 ***Are you collecting the Social Security Number of any subjects for any purpose?***
No

X.4 ***How will information/data be collected and stored for this study (check all that apply):***

- Biologic samples (blood draws, check swabs, saliva samples, tissue samples, etc.) - Blood samples will be sent to the UIHC pathology lab for analyses. Remaining biological specimens, such as blood, will be labeled with subject code, date collected and IRB protocol number and transported to the PIs laboratory (118 PSRB) in a secure unbreakable biohazard container. Samples will be stored in the PIs laboratory in a -80C freezer in 111 PSRB. All samples will be labeled with date collected and subject ID code only. No personal identifiable information will be labeled on the sample. Only the PI and her research staff will have access to the samples.
  - Name - Anna Reid-Stanhewicz
  - Title - Assistant Professor
  - University Job Classification - Faculty/Staff
- Electronic records (computer files, electronic databases, etc.) - Electronic records (computer files, electronic databases, etc.) - Data will be entered using subject ID code into the ICTS REDCap web-based database application that is password protected. No personal identifiable data will be entered. Only research staff on the IRB approved study will be allowed access this database. The ICTS REDCap staff are responsible for maintaining security of the data. Some data using subject ID code will also be entered into a Microsoft Excel and SPSS datasheets that will be kept in a shared server for CLAS that is password protected. Only research staff on the IRB approved study will have access to the folder the study on the server.
  - Name - Bryan Ringen
  - Title - IT Support Consultant
  - University Job Classification - Faculty/Staff
- Paper/hard copy records (hard copy surveys, questionnaires, case report forms, pictures, etc.) - Most of the data are coded and do not contain personal identifying information. Some data will temporarily contain the subject's name, address, and/or telephone number (Phone Interview Form). Documents allowing identification of participants do not leave our labs and are only available to authorized persons. Only authorized personnel may access the lab computer. Data forms containing identifiable information are shredded when no longer needed (within 5 years after publication of results). We keep data in the laboratory in locked cabinets, the password-protected folder on the secured University of Iowa RDSS server, and on password-protected computers maintained in locked laboratory rooms. Only authorized personnel have access. Coded data shared with unauthorized persons cannot be traced to individuals. The list linking code numbers to participants is not shared with unauthorized persons and destroyed when the project is completed and within 5 years of publication of the data. Screening data from subjects who are not accepted into the study are shredded when the project ends. Subjects may give permission to have their contact information retained in the investigator's secured files if they wish to be considered for participation in future studies. After we complete the study, we remove all identifiers from the data and store the data indefinitely. Individual data may be used without identifying the subject to illustrate representative responses. Any hard paper copy of subject lists or data will be kept in a locked cabinet in the locked office of the PI. Subjects will be assigned a code for the study. Files will be labeled with this code for confidentiality. Study data including laser-Doppler flowmetry data will be stored on password protected computer hard-drives and password protected server folders which are only accessible to research team members via password. Subject confidentiality will be maintained in all presentations and publications and information/records pertaining to subject participation in the research project will not be released without prior authorization by the subjects.

X.5 ***Do the confidentiality protections indicated above allow only members of the research team to access the data/specimens?***
No

X.6 ***Describe***
The -80C freezer in 111 PSRB is a shared wet lab space. The door to this lab space is kept locked but those with keys to the shared lab space would be able to enter the room. Individuals with key access to 111 PSRB include:
Nathaniel Jenkins, PhD
Mark Flores, BS
Laura Schwager, BA

X.7 ***Does your study meet the NIH criteria for a Certificate of Confidentiality or will you be applying for Certificate of Confidentiality?***
Yes

X.8 ***If yes, provide rationale:***
Per Section 2012 of the 21st Century Cures Act as implemented in the 2017 NIH Certificates of Confidentiality Policy, all ongoing or new research funded by NIH as of December 13, 2016 that is collecting or using identifiable, sensitive information is automatically issued a CoC.

XI. Data Analysis

XI.1 ***Describe the analysis methods you will use, including, if applicable, the variables you will analyze***
The primary outcome variable will be cutaneous vascular conductance, which is calculated from laser-Doppler flux/mean arterial pressure. Three-way repeated measures ANOVA (treatment, MD site, dose) will be used to examine systemic treatment differences (losartan vs. placebo) and local

microdialysis treatment differences across the doses of pharmacological stimuli. Appropriate post-hoc analyses with corrections for multiple comparisons will be performed when main effects are identified, including potential confounding variables as covariates. Paired t-tests will be used to examine systemic treatment differences on pulse wave velocity.

XI.2 ***Provide the rationale or power analysis to support the number of subjects proposed to complete this study.***
Based on previous data examining chronic pharmacological intervention strategies we calculate that 10 subjects will be necessary to detect an effect size of 15% between microdialysis sites (1 sample RMANOVA, power=0.08, a=0.05). Based on the duration of the intervention and prior experience, we anticipate that ~5 subjects will screen but not be eligible and of those that enroll, ~5 will drop out of the intervention, therefore, we suggest a total sample of 20 subjects.

XII. Future Research

XII.1 ***Do you wish to keep any information about subjects involved with this research project so that members of the current research team may contact them in the future for your own research projects?***
Yes

XII.2 ***Do you wish to keep any information about subjects involved with this research project so that other researchers may contact them for future research?***
No

XII.3 ***List the data or information you will keep:***
Name
Race/ethnicity
Age
Pregnancy history
Contact information (mailing address, e-mail address, phone number)

XII.4 ***Does this project involve storing any data, tissues or specimens for future research?***
Yes – contribution for future use is optional

XII.5 ***Describe how you will keep track of those who consent to future use and those who do not and how you will prevent future use for those who do not consent.***
We will keep track of who consents to future use on our enrollment log.