Methods

Subjects: This multicenter study included three sites. Rochester, Minnesota enrolled non-Hispanic whites. Jackson, Mississippi enrolled non-Hispanic blacks. These two sites recruited sibships in which at least two siblings developed hypertension before age 60. Starr County, Texas enrolled Hispanics. To avoid potential confounding due to the high prevalence of diabetes among Mexican Americans, this site recruited sibships in which at least two siblings had diabetes. The study was completed in two phases (Phase 1: 1996-2000; Phase 2: 2000-2004). [1-3] 5,424 participants completed Phase 1, and 72% returned for Phase 2. Phase 2 also included 28 additional siblings or spouses of Phase I participants who were recruited in Rochester. The present analysis was limited to Phase 2 participants, as the pregnancy history questionnaire was only included in Phase 2. The study included questionnaires, a physical exam and a fasting blood sample.

Questionnaires: Questionnaires were administered by a trained interviewer. Phase 2 included questionnaires regarding personal and family medical history. Participants provided information about whether they had experienced a coronary heart disease event or stroke, or been diagnosed with hypertension or diabetes. For each outcome that participants had experienced, they reported the year in which the outcome was first diagnosed or occurred. Women also completed a validated questionnaire concerning hypertension in pregnancy.[4] Women were asked 'Have you had at least one pregnancy lasting more than 6 months?' Women who responded 'yes' were asked how many pregnancies they had, whether they developed hypertension in any pregnancy lasting more than 6 months, and whether they developed preeclampsia in any pregnancy lasting

more than 6 months. Women who reported hypertension or preeclampsia during any pregnancy were asked to report the number of pregnancies affected by hypertension or preeclampsia. The pregnancy history questionnaire had an 80% sensitivity and 90% specificity for the determination of preeclampsia among women in Rochester, Minnesota.[4]

Physical Examination: Blood pressure, weight and height were measured using a standardized protocol. Blood pressure was measured with an automated oscillometric device. Height was measured with the subject standing with her heels together, without shoes, against a vertically mounted ruler. Venipuncture was performed after an overnight fast (≥8 hours). Serum triglyceride, total cholesterol and HDL cholesterol concentrations were measured on a Hitachi 911 Chemistry Analyzer (Roche Diagnostics, Indianapolis, Indiana, USA).

Statistical analysis: Continuous data are expressed as mean ± standard deviation. Categorical data are presented as absolute numbers with percentages. Unadjusted and adjusted prevalences for coronary heart disease events, stroke, cardiovascular disease (coronary heart disease event or stroke) and hypertension are shown. Prevalences were adjusted for participant characteristics that differed between the groups that were being compared. Cox proportional hazard models were conducted with age as the time scale. In the case of an event, age at onset of the event was used; while for a censored observation in the absence of an event current age was used as the time variable. In case of multiple events, age at which the first event was reported was used to calculate outcome free survival. The effects of a personal or sibling history of hypertension in pregnancy on disease risk in men and women were determined using hazard ratios (HRs) presented with 95% confidence intervals (CIs) after adjusting for potential confounders (see

detailed descriptions below). Participants were recruited through different networks; therefore regression models included hypertension in pregnancy x network interaction. This interaction was not significant and was removed from the final models. To examine differences in the time to each event, Kaplan-Meier curves were constructed and groups were compared using the Log-Rank test. Statistical analyses were performed using SPSS (SPSS for Windows, release 21.0, SPSS, Chicago, IL). In all tests, $p<0.05$ was considered to be statistically significant.

Effect of a personal history of hypertension in pregnancy (within-family comparisons): Cox regression models with clustering by sibship were used for within family comparisons. The effects of a personal history of hypertension in pregnancy on future disease risk were determined using hazard ratios (HRs) presented with 95% confidence intervals (CIs) after adjusting for participant characteristics that differed between the study groups (BMI, diabetes). The final analyses combined nulliparous and parous sisters. Results were not different in a sensitivity analysis that excluded nulliparous sisters. For the outcome of hypertension, women who developed hypertension prior to or during the year of their first hypertensive pregnancy were excluded from the analysis.

Effect of a sibling history (Between-family comparisons): Cox regression models were also performed for across family comparisons. The effects of a sibling history of hypertension in pregnancy on future disease risk were determined using hazard ratios (HRs) presented with 95% confidence intervals (CIs) after adjusting for participant characteristics that differed between the study groups (BMI, smoking), number of parous sisters in the family and family history of the outcome. Due to potential influence of intra-familial correlation of outcomes when comparing

sibships across families (participants with vs. without a sister who had a HPD) separate analysis was performed by using the complex re-sampling method, that leaves out each family, one at a time, in turn. For the outcome of hypertension, siblings of women who developed hypertension prior to or during the year of their first hypertensive pregnancy were excluded from the analysis. Family history was defined as a self-reported maternal or paternal history of the outcome. In the analyses presented in the paper, family history was not adjusted in the case of disagreement between siblings. A sensitivity analysis was performed to determine whether results were different when family history was adjusted such that all siblings in the family were considered to have a family history if any sibling reported a maternal or paternal history of the outcome.

References

1. Multi-center genetic study of hypertension: The Family Blood Pressure Program (FBPP). *Hypertension,* 39**:** 3-9, 2002.
2. Garovic, VD, Bailey, KR, Boerwinkle, E, Hunt, SC, Weder, AB, Curb, D, Mosley, TH, Jr., Wiste, HJ, Turner, ST: Hypertension in pregnancy as a risk factor for cardiovascular disease later in life. *Journal of hypertension,* 28**:** 826-833, 2010.
3. Weissgerber, TL, Turner, ST, Bailey, KR, Mosley, TH, Jr., Kardia, SL, Wiste, HJ, Miller, VM, Kullo, IJ, Garovic, VD: Hypertension in pregnancy is a risk factor for peripheral arterial disease decades after pregnancy. *Atherosclerosis,* 229**:** 212-216, 2013.
4. Diehl, CL, Brost, BC, Hogan, MC, Elesber, AA, Offord, KP, Turner, ST, Garovic, VD: Preeclampsia as a risk factor for cardiovascular disease later in life: validation of a preeclampsia questionnaire. *Am J Obstet Gynecol,* 198**:** e11-13, 2008.