Euglycemic Clamp Insulin Sensitivity and Longitudinal Systolic Blood Pressure: Role of Sex

John R. Petrie, Muhammad Omar Malik, Beverley Balkau, Colin G. Perry, Kurt Højlund, Zoltan Pataky, John Nolan, Ele Ferrannini and Andrea Natali for the RISC Investigators

_Hypertension._ 2013;62:404-409; originally published online June 3, 2013; doi: 10.1161/HYPERTENSIONAHA.111.00439

_Hypertension_ is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
Copyright © 2013 American Heart Association, Inc. All rights reserved.
Print ISSN: 0194-911X. Online ISSN: 1524-4563

The online version of this article, along with updated information and services, is located on the World Wide Web at:
http://hyper.ahajournals.org/content/62/2/404

Data Supplement (unedited) at:
http://hyper.ahajournals.org/content/suppl/2013/06/03/HYPERTENSIONAHA.111.00439.DC1.html
http://hyper.ahajournals.org/content/suppl/2013/06/03/HYPERTENSIONAHA.111.00439.DC2.html

Permissions: Requests for permissions to reproduce figures, tables, or portions of articles originally published in _Hypertension_ can be obtained via RightsLink, a service of the Copyright Clearance Center, not the Editorial Office. Once the online version of the published article for which permission is being requested is located, click Request Permissions in the middle column of the Web page under Services. Further information about this process is available in the Permissions and Rights Question and Answer document.

Reprints: Information about reprints can be found online at:
http://www.lww.com/reprints

Subscriptions: Information about subscribing to _Hypertension_ is online at:
http://hyper.ahajournals.org/subscriptions/
Insulin Sensitivity and Blood Pressure

Euglycemic Clamp Insulin Sensitivity and Longitudinal Systolic Blood Pressure
Role of Sex

John R. Petrie, Muhammad Omar Malik, Beverley Balkau, Colin G. Perry, Kurt Højlund, Zoltan Pataky, John Nolan, Ele Ferrannini, Andrea Natali; for the RISC Investigators

Abstract—Insulin resistance may be an independent risk factor for the development of hypertension, but change in blood pressure (BP) over time has not been adequately studied in healthy individuals fully characterized for insulin sensitivity. In the Relationship between Insulin Sensitivity and Cardiovascular disease (RISC) study, we measured insulin sensitivity (M/I) using the euglycemic clamp technique in 1073 healthy European adults (587 women, 486 men) aged 30 to 60 years followed up 3 years later. Systolic BP (SBP) at baseline was higher in insulin-resistant women (ie, those in the low sex-specific M/I tertile) compared with those in the intermediate (P<0.001) or high tertiles (P=0.06; mean±SD: 117±13, 111±12, 114±12 mm Hg, respectively). It did not differ across M/I tertiles in men. After adjustment for age, body mass index, baseline SBP, and other covariates, low insulin sensitivity (M/I) predicted a longitudinal rise in SBP in women but not in men; M/I was not associated with change in diastolic BP. SBP rose over time in both sexes and within all M/I tertiles (P<0.05), except in women with high insulin sensitivity. Therefore, in women (but not in men), low insulin sensitivity was associated with higher SBP at 3 years, and high insulin sensitivity was associated with a lower rise in SBP over time. (Hypertension. 2013;62:404-409.) ● Online Data Supplement

Key Words: blood pressure ■ hypertension ■ insulin ■ insulin resistance ■ sex

Hyperinsulinemia1 and underlying insulin resistance are associated with essential hypertension,2 but their exact role in pathophysiology remains unclear. In the Framingham Offspring study,3 an insulin sensitivity index estimated from fasting and postload insulin and glucose levels4 independently predicted blood pressure (BP) tracking during 4 years in non-obese middle-aged men and women.5,6

The euglycemic clamp technique, requiring intravenous insulin infusion with real-time glucose monitoring, is the acknowledged gold standard for the assessment of insulin sensitivity.7 However, because it is invasive and labor-intensive, it has not been applied in adequately sized cohorts at risk of hypertension. Longitudinal studies have instead used fasting insulin concentrations, homeostasis model assessment (HOMA; calculated from fasting insulin and glucose concentrations), and insulin sensitivity index (calculated from fasting and postload insulin and glucose levels).4,8 Most,9-22 but not all,23-27 have reported a relationship between insulin resistance and future BP rise or the development of hypertension.

Although these surrogate measures correlate with clamp-measured insulin sensitivity,28 they depend on insulin immunoassays and do not account for body mass or body composition. Some previous studies in adults have used more direct physiological methods but have been either small29 or cross-sectional.30,31

Having previously published cross-sectional euglycemic clamp data from the Relationship between Insulin Sensitivity and Cardiovascular disease (RISC) cohort (n=1384),32 we now report the longitudinal relationship between baseline insulin sensitivity and BP during 3 years.

Methods

The RISC study is a large euglycemic clamp–based prospective study of the association between insulin resistance and atherosclerotic disease.33 The RISC investigators and recruiting centers are listed in full in the online-only Data Supplement. The protocol was approved by Institutional Review Committees at all study centers, and the participants gave informed consent. All clinical assessments were performed according to the principles of the Declaration of Helsinki.

Participants were clinically healthy individuals aged 30 to 60 years recruited from 18 centers in 13 European countries. Exclusion criteria included presence of (or treatment for) cardiovascular disease, diabetes mellitus, dyslipidemia, or any other chronic disease. Of the 1563 participants screened, 1384 healthy individuals had a

Received November 9, 2012; first decision December 16, 2012; revision accepted May 12, 2013.
From the University of Glasgow, Glasgow, United Kingdom (J.R.P., M.O.M., C.P.); Khyber Medical University Peshawar, Peshawar, Pakistan (M.O.M.); INSERM U1018, Villejuif, France (B.B.); University Paris Sud 11, UMR 1018, Villejuif, France (B.B.); Odense University Hospital, Odense, Denmark (K.H.); University Hospital of Geneva, Switzerland (Z.P.); Steno Diabetes Center, Copenhagen, Denmark (J.N.); and the Department of Internal Medicine, University of Pisa, Pisa, Italy (E.F., A.N.).
The online-only Data Supplement is available with this article at http://hyper.ahajournals.org/lookup/suppl/doi:10.1161/HYPERTENSIONAHA.111.00439/-/DC1.
Correspondence to John R. Petrie, Professor of Diabetic Medicine, BHF Glasgow Cardiovascular Research Centre, University of Glasgow, 126 University Place, Glasgow G12 8TA, United Kingdom. E-mail john.petrie@glasgow.ac.uk
© 2013 American Heart Association, Inc.
Hypertension is available at http://hyper.ahajournals.org
DOI: 10.1161/HYPERTENSIONAHA.111.00439

Downloaded from http://hyper.ahajournals.org/ at Bibliothèque Faculte Medecine Geneve on April 1, 2014
euglycemic clamp at baseline; of these, 1073 (587 women and 486 men) had complete data at 3-year follow-up after exclusion of those who had developed diabetes mellitus and symptomatic cardiovascular disease (n=21).

**BP and Physical Examination**

BP was measured sitting after 5-minute rest by trained study nurses (OMRON 705CP, Hamburg, Germany) in triplicate, according to a standard protocol: the median of these readings was used in this analysis. Median systolic BP (SBP) ≥140 mm Hg and median diastolic BP (DBP) ≥90 mm Hg at follow-up were taken to indicate hypertension.33 Participants who had been identified and commenced on treatment in routine care (n=46) were classified as hypertensive.

Height was measured by stadiometer. Body weight, percentage of body fat, and lean body mass (ffm) were evaluated by bioimpedance (TBF-300, Tanita International Division, United Kingdom).

**Insulin Sensitivity**

Insulin sensitivity was measured by a standard hyperinsulinemic euglycemic clamp technique as previously described, after central training of site staff.33 Target plasma glucose concentration was between 4.5 and 5.5 mmol/L; insulin was infused at a rate of 240 pmol-min⁻¹·m⁻². Plasma glucose was measured at 5- to 10-minute intervals to ensure it remained within 0.8 mmol/L (±15%) of target glucose concentration. The steady-state period (for calculation of insulin sensitivity) was between 80 and 120 minutes. The glucose infusion rate (M value) was expressed in mg·min⁻¹·lbm(kg)⁻¹. Insulin sensitivity was expressed as M/I (μmol·min⁻¹·kglbm⁻¹·mmol/L⁻¹), with the M value divided by steady-state insulin.

**Biological Samples**

Total cholesterol, high-density lipoprotein cholesterol, and triglycerides were assayed by enzymatic colorimetric assay (Roche), and low-density lipoprotein cholesterol was calculated by the Friedewald formula. A 75-g oral glucose tolerance test was used to exclude participants with diabetes mellitus at inclusion (or developing diabetes mellitus during follow-up), with glucose assayed centrally by the glucose oxidase method (Cobas Integra, Roche).

**Other Measures**

Physical activity was measured during 1 week by Actigraph accelerometer (version AM7164-2.2, CSA Inc, Florida) expressed as average number of counts per minute.36 Women were classified as postmenopausal if their last menstrual period was ≥12 months before treatment (TBF-300, Tanita International Division, United Kingdom). BP values for individuals who had been commenced on antihypertensive medication between baseline and follow-up.34 The relationship between log M/I and SBP at Year 3 adjusted for baseline SBP was used in preference to the unadjusted relationship with change in SBP, given that baseline BP and change in BP are usually highly correlated.15 The relationship was curvilinear when adjusted for study center and age only, but this was no longer the case when baseline SBP was added to the model. β-coefficients for insulin sensitivity are shown along with R² for the coefficient of determination for the model. Binary logistic regression was used to assess prediction of hypertension defined according to ESH/JNCVII (European Society of Hypertension/Seventh Report of the Joint National Committee on Prevention, Detection, Evaluation and Treatment of High Blood Pressure) as ≥140/≥90 mm Hg35 (or by its treatment). SPSS version 18 was used in all analyses.

**Results**

At 3 years, BP had reached a diagnostic threshold for hypertension in 11.6% of all participants (n=125; 75 men, 50 women). A further 4.3% (n=46; 23 men, 23 women) had been commenced on antihypertensive treatment in routine care, that is, 16.0% (n=171; 98 men, 73 women) of all participants had developed incident hypertension.

Insulin concentrations achieved at steady state (mean±SD) were 416±111 pmol/L in men and 406±112 pmol/L in women. Men had lower insulin sensitivity (M/I) than women (Table 1); statistically significant differences were also noted between the sexes for all measurements, except smoking status and physical activity.

In univariate analyses (Table 2), change in both systolic and DBP correlated in the expected manner with baseline values in both sexes. Change in both systolic and DBP from baseline was correlated with insulin sensitivity (log M/I) in women but not in men. Weight correlated with change in diastolic, but not SBP, in both sexes. Other correlations were of borderline statistical significance.

When baseline SBP was expressed according to sex-specific tertiles of insulin sensitivity (M/I; mean±SD; Figure; for cut off values see figure legend), there were no differences between tertiles among men (123±11 [low]; 121±11 [intermediate]; 123±10 mm Hg [high]). However, women with low baseline M/I tended to have higher baseline SBP: 117±13 mm Hg (low), 111±12 mm Hg (intermediate), 114±12 mm Hg (high; low versus intermediate, 6.0 [95% confidence interval (CI), 3.0–9.0] mm Hg, P<0.001; low versus high 3.0 [95% CI, −0.1 to 5.9] mm Hg, P=0.06).

During 3 years of follow-up, SBP increased from baseline in all tertiles for men (P<0.05; Figure). In women, SBP rose in those with low and intermediate M/I (P<0.05), but no change was observed in the high insulin sensitivity tertile (114±12 versus 114±14 mm Hg; P=0.791; 0.2 [95% CI, −1.8 to 1.4]). Comparing unadjusted 3-year SBP between insulin sensitivity tertiles within each sex (Tukey post hoc testing), no statistically significant differences were observed in men: (127±13 [low]; 126±13 [intermediate], 126±14 mm Hg [high]). However, in women, 3-year SBP in the low M/I tertile (121±16 mm Hg) was significantly higher than in the intermediate (116±16 mm Hg; P=0.001; 5.6 [95% CI, 1.93–9.26]) and high (114±14 mm Hg) M/I tertiles (P<0.001; 7.1 [95% CI, 3.47–10.79]). Test for trend for 3-year SBP across the tertiles was significant in women (P<0.001) but not in men.
When these analyses were repeated, excluding individuals on antihypertensive treatment, similar results were obtained (data not shown). When key covariates (including recruitment center, age, baseline SBP, BMI, change in BMI, lipid profile, smoking status, and fasting glucose) were included in multiple regression analyses (Table 3), low insulin sensitivity (log M/I) significantly and independently predicted SBP at 3 years in women ($\beta$ = −0.214; $P < 0.001$) but not in men. M/I was not a predictor of the development of hypertension, in either sex, after adjustment for baseline SBP, age, and BMI (data not shown), although the trend test across the M/I tertiles was significant for the development of hypertension in women ($P < 0.01$) but not in men ($P = 0.260$). There was no relationship between insulin sensitivity and SBP in either sex when HOMA was substituted for Log M/I as an independent variable (data not shown). Insulin sensitivity did not predict DBP longitudinally (data not shown).

**Discussion**

In this analysis of 1073 healthy European adults, low insulin sensitivity measured using a robust euglycemic clamp technique predicted rise in SBP at 3 years in women but not in men. SBP was higher at baseline in women with low insulin sensitivity than that in those with intermediate or high insulin sensitivity. It increased during 3 years in all groups studied, except in women with high baseline insulin sensitivity. Insulin sensitivity predicted change in SBP independently of key covariates (including age, baseline BP, BMI, and change in BMI) in women only. These findings offer new insights into the relationships between metabolic factors and BP by sex, clarifying and extending cross-sectional data we have previously reported from the RISC cohort.

The most comprehensive previous study on this topic was the Framingham Offspring study in which insulin sensitivity was assessed in 1933 healthy adults using an insulin sensitivity index based on fasting and postload insulin and glucose levels. In this report, insulin sensitivity (expressed categorically in sex-specific quartiles and stratified by age) was independently associated with BP over time in younger, leaner individuals of both sexes but not in those who were older, overweight, or obese. A unique feature of the present analysis is that it is based on data from a large number of individuals undergoing a standard euglycemic clamp. Insulin sensitivity was directly derived from the glucose infusion rate during steady-state euglycemia adjusted only for centrally measured insulin concentrations, and lean body mass was measured using a standard device. Only 2 previous investigations into the longitudinal relationship between insulin sensitivity and BP in adults have incorporated direct measures of insulin sensitivity, and only 1 investigation used the euglycemic clamp. One of these studies was relatively small (n=54) and in men only: no effect was demonstrated. The other used a modified frequently sampled intravenous glucose tolerance test in a triethnic population (n=840) and reported a modest protective association of insulin sensitivity on the risk of hypertension.

Other longitudinal studies reporting a relationship between insulin sensitivity and BP have been based on surrogate
measures of insulin resistance, including either fasting insulin\textsuperscript{10–15,17,21,22} or fasting insulin and glucose concentrations (HOMA).\textsuperscript{18,19} They have been performed in a variety of populations (Scandinavian\textsuperscript{11,15,22}; Mexican-American\textsuperscript{16,17}; Japanese individuals\textsuperscript{13,14}), with some including only men\textsuperscript{10,11,19,25,29} but none only women. Some studies of reasonable size and duration have reported no relationship between insulin sensitivity and BP\textsuperscript{9,23,24,26,27} after adjustment.

Table 2. Pearson Correlations (\(r\)) Between Baseline Characteristics and Change (\(\Delta\)) in Systolic and Diastolic BP During Three Years

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>(\Delta)SBP Male</th>
<th>(\Delta)SBP Female</th>
<th>(\Delta)DBP Male</th>
<th>(\Delta)DBP Female</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, y</td>
<td>0.162‡</td>
<td>0.027</td>
<td>0.074</td>
<td>−0.068</td>
</tr>
<tr>
<td>Baseline SBP, mmHg</td>
<td>−0.293‡</td>
<td>−0.273‡</td>
<td>−0.162‡</td>
<td>−0.163‡</td>
</tr>
<tr>
<td>Baseline DBP, mmHg</td>
<td>−0.293‡</td>
<td>−0.196‡</td>
<td>−0.340‡</td>
<td>−0.339‡</td>
</tr>
<tr>
<td>Waist, cm</td>
<td>0.056</td>
<td>0.055</td>
<td>0.067</td>
<td>0.008</td>
</tr>
<tr>
<td>Weight, kg</td>
<td>0.077</td>
<td>0.073</td>
<td>0.113*</td>
<td>0.134†</td>
</tr>
<tr>
<td>BMI, kg/m(^2)</td>
<td>0.034</td>
<td>0.081*</td>
<td>0.086</td>
<td>0.100*</td>
</tr>
<tr>
<td>Fat free mass, kg</td>
<td>0.092</td>
<td>0.050</td>
<td>0.079</td>
<td>0.162‡</td>
</tr>
<tr>
<td>Fat mass, kg</td>
<td>0.040*</td>
<td>0.074</td>
<td>0.110*</td>
<td>0.103*</td>
</tr>
<tr>
<td>Glucose, mmol/L</td>
<td>−0.008</td>
<td>0.016</td>
<td>−0.005</td>
<td>−0.037</td>
</tr>
<tr>
<td>Total cholesterol, mmol/L</td>
<td>0.029</td>
<td>0.076</td>
<td>−0.041</td>
<td>−0.012</td>
</tr>
<tr>
<td>LDL cholesterol, mmol/L</td>
<td>0.061</td>
<td>0.080</td>
<td>−0.044</td>
<td>−0.017</td>
</tr>
<tr>
<td>HDL cholesterol, mmol/L</td>
<td>−0.153†</td>
<td>−0.009</td>
<td>−0.100*</td>
<td>−0.007</td>
</tr>
<tr>
<td>TG, mmol/L</td>
<td>0.075</td>
<td>0.016</td>
<td>0.058</td>
<td>0.006</td>
</tr>
<tr>
<td>Insulin sensitivity, log M/I</td>
<td>−0.054</td>
<td>−0.132†</td>
<td>−0.045</td>
<td>−0.110†</td>
</tr>
<tr>
<td>Smoker, %</td>
<td>0.019</td>
<td>−0.004</td>
<td>−0.010</td>
<td>−0.022</td>
</tr>
<tr>
<td>Alcohol, g/wk</td>
<td>0.021</td>
<td>0.044</td>
<td>0.012</td>
<td>0.057</td>
</tr>
<tr>
<td>Physical activity, counts/min worn</td>
<td>−0.014</td>
<td>0.000</td>
<td>−0.024</td>
<td>−0.103*</td>
</tr>
<tr>
<td>Creatinine, µmol/L</td>
<td>−0.041</td>
<td>−0.031</td>
<td>0.052</td>
<td>0.080</td>
</tr>
<tr>
<td>eGFR, mL/min per 1.73 m(^2)</td>
<td>0.055</td>
<td>0.030</td>
<td>−0.035</td>
<td>−0.067</td>
</tr>
</tbody>
</table>

BMI indicates body mass index; BP, blood pressure; DBP, diastolic blood pressure; eGFR, estimated glomerular filtration rate; HDL, high-density lipoprotein; LDL, low-density lipoprotein; M/I, glucose metabolised/insulin; SBP, systolic blood pressure; and TG, triglyceride.

Table 3. Standardized β Coefficients for Predicting Systolic BP at Three-Year Follow-Up From Log M/I as Independent Variable (With Various Adjustment Factors)

<table>
<thead>
<tr>
<th>Model</th>
<th>Adjustment Factors</th>
<th>Men (n=486)</th>
<th>Women (n=587)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>R(^2)</td>
<td>(\beta)</td>
</tr>
<tr>
<td>1</td>
<td>…</td>
<td>0.005</td>
<td>−0.069</td>
</tr>
<tr>
<td>2</td>
<td>Center and age</td>
<td>0.147</td>
<td>−0.049</td>
</tr>
<tr>
<td>3</td>
<td>Model 2 + baseline systolic BP</td>
<td>0.365</td>
<td>−0.034</td>
</tr>
<tr>
<td>4</td>
<td>Model 3 + BMI</td>
<td>0.368</td>
<td>−0.007</td>
</tr>
<tr>
<td>5</td>
<td>Model 4 + %change BMI</td>
<td>0.385</td>
<td>−0.003</td>
</tr>
<tr>
<td>6</td>
<td>Model 5 + baseline eGFR</td>
<td>0.386</td>
<td>−0.004</td>
</tr>
<tr>
<td>7</td>
<td>Model 6 + glucose, Chol, LDL, HDL, log TG</td>
<td>0.397</td>
<td>0.030</td>
</tr>
<tr>
<td>8</td>
<td>Model 7 + baseline HRT &amp; OCP use</td>
<td>0.397</td>
<td>0.030</td>
</tr>
<tr>
<td>9</td>
<td>Model 8 + smoking</td>
<td>0.398</td>
<td>0.031</td>
</tr>
<tr>
<td>10</td>
<td>Model 9 + phys. activity</td>
<td>0.396</td>
<td>0.026</td>
</tr>
<tr>
<td>11</td>
<td>Model 10 + log alcohol</td>
<td>0.396</td>
<td>0.026</td>
</tr>
</tbody>
</table>

BMI indicates body mass index; BP, blood pressure; eGFR, estimated glomerular filtration rate; HDL, high-density lipoprotein; HRT, hormone replacement therapy; LDL, low-density lipoprotein; M/I, glucose metabolised/insulin; OCP, oral contraceptive pill; and TG, triglyceride.
protocol among participating centers; central training of study personnel with focus on quality control; and accurate measurement of BP using the same standard and validated device in all centers. We acknowledge that the low proportion of cases that developed incident hypertension (16%) limited the power of our analysis to address a more clinically relevant outcome measure. Moreover, given the invasive nature of assessments, we were unable to use a truly population-based sampling method. Because only European centers were included, with very few participants representing ethnic minorities, it cannot be assumed that our findings can be generalized to other populations.

**Perspectives**

These prospective data from the Europe-wide RISC cohort of healthy adults indicate that low insulin sensitivity measured using the euglycemic clamp technique is an independent predictor of longitudinal change in BP over time in women but not in men. Women with high insulin sensitivity may be protected against rise in SBP over time. The physiological basis for the sex difference we report in the RISC cohort and its implications for the role of insulin resistance in the pathophysiology of hypertension remain uncertain. Further insights may be gained by further follow-up of the cohort.

**Sources of Funding**

The Relationship between Insulin Sensitivity and Cardiovascular disease (RISC) study was supported by EU Framework Program V grant QLG1-CT-2001-01252 with additional funding from AstraZeneca (Sweden). RISC is run by the European Group for the study of Insulin Resistance (EGIR), which is supported by an unrestricted research grant from Merck-Serono. J. Petrie received support for this analysis from a Travelling Fellowship from the Royal College of Physicians and Surgeons of Glasgow.

**Disclosures**

None.

**References**


Novelty and Significance

**What Is New?**
- The RISC study is the longest longitudinal study of the relationship between euglycemic clamp insulin resistance and BP in healthy men and women.

**What Is Relevant?**
- Women with high insulin sensitivity may be protected against rise in systolic BP over time.

**Summary**
Low euglycemic clamp-measured insulin sensitivity independently predicted systolic BP at 3 years in women but not in men.
ONLINE SUPPLEMENT

For:

EUGLYCEMIC CLAMP INSULIN SENSITIVITY AND LONGITUDINAL SYSTOLIC BLOOD PRESSURE:

ROLE OF GENDER

John R Petrie, MD, PhD; Muhammad Omar Malik, MD; Beverley Balkau, PhD; Colin G Perry, MD, PhD; Kurt Højlund, MD; Zoltan Pataky, MD; John Nolan, MD; Ele Ferrannini, MD, PhD; Andrea Natali, MD, PhD

for the RISC Investigators

From the University of Glasgow, United Kingdom (J.R.P., M.O.M., C.P.); Khyber Medical University Peshawar, Pakistan (M.O.M); INSERM U1018, Villejuif, France (B.B.); University Paris Sud 11, UMRS 1018, Villejuif, France (B.B.); Odense University Hospital, Odense, Denmark (K.H.); University Hospital of Geneva, Switzerland (Z.P.); Steno Diabetes Center, Copenhagen, Denmark (J.N.); and the Department of Internal Medicine, University of Pisa, Pisa, Italy (E.F., A.N.)

Short title: Insulin sensitivity, gender and blood pressure

Corresponding Author:

Professor John R Petrie BSc, MBChB, PhD, FRCP(Ed), FRCPSG.
Professor of Diabetic Medicine
BHF Glasgow Cardiovascular Research Centre
University of Glasgow
126 University Place
Glasgow G12 8TA
Tel +44(0)141 330 3325
Fax +44(0)141 330 6972
E-mail john.petrie@glasgow.ac.uk
RISC investigators and recruiting centres:

**Amsterdam, The Netherlands:** RJ Heine, J Dekker, S de Rooij, G Nijpels, W Boorsma

**Athens, Greece:** A Mitrakou, S Tournis, K Kyriakopoulou, P Thomakos

**Belgrade, Serbia:** N Lalic, K Lalic, A Jotic, L Lukic, M Civcic

**Dublin, Ireland:** J Nolan, TP Yeow, M Murphy, C DeLong, G Neary, MP Colgan, M Hatunic

**Frankfurt, Germany:** T Konrad, H Böhles, S Fuellert, F Baer, H Zuchhold

**Geneva, Switzerland:** A Golay, E Harsch Bobbioni, V. Barthassat, V. Makoundou, TNO Lehmann, T Merminod

**Glasgow, Scotland:** JR Petrie, CG Perry, F Neary, C MacDougall, K Shields, L Malcolm

**Kuopio, Finland:** M Laakso, U Salmenniemi, A Aura, R Raisanen, U Ruotsalainen, T Sistonen, M Laitinen, H Saloranta

**London, England:** SW Coppack, N McIntosh, J Ross, L Pettersson, P Khadobaksh

**Lyon, France:** M Laville, F. Bonnet (now Rennes), A Brac de la Perriere, C Louche-Pelissier, C Maitrepierre, J Peyrat, S Beltran, A Serusclat

**Madrid, Spain:** R. Gabriel, EM Sánchez, R. Carraro, A Friera, B. Novella

**Malmö, Sweden (1):** P Nilsson, M Persson, G Östling, (2): O Melander, P Burri

**Milan, Italy:** PM Piatti, LD Monti, E Setola, E Galluccio, F Minicucci, A Colleluori

**Newcastle-upon-Tyne, England:** M Walker, IM Ibrahim, M Jayapaul, D Carman, C Ryan, K Short, Y McGrady, D Richardson

**Odense, Denmark:** H Beck-Nielsen, P Staehr, K Hojlund, V Vestergaard, C Olsen, L Hansen

**Perugia, Italy:** GB Bolli, F Porcellati, C Fanelli, P Lucidi, F Calcinaro, A Saturni

**Pisa, Italy:** E Ferrannini, A Natali, E Muscelli, S Pinnola, M Kozakova, A Casolaro, BD Astiarraga

**Rome, Italy:** G Mingrone, C Guidone, A Favuzzi, P Di Rocco
Vienna, Austria: C Anderwald, M Bischof, M Promintzer, M Krebs, M Mandl, A Hofer, A Luger, W Waldhäusl, M Roden

**Project Management Board:** B Balkau (Villejuif, France), SW Coppack (London, England), JM Dekker (Amsterdam, The Netherlands), E Ferrannini (Pisa, Italy), A Golay (Geneva, Switzerland), A Mari (Padova, Italy), A Natali (Pisa, Italy), JR Petrie (Glasgow, Scotland), M Walker (Newcastle, England)

**Core laboratories and reading centres:**

**Lipids** Dublin, Ireland: P Gaffney, J Nolan, G Boran

**Hormones** Odense, Denmark: C Olsen, L Hansen, H Beck-Nielsen

**Albumin:creatinine** Amsterdam, The Netherlands: A Kok, J Dekker

**Genetics** Newcastle-upon-Tyne, England: S Patel, M Walker

**Stable isotope laboratory** Pisa, Italy: A Gastaldelli, D Ciociaro

**Adiponectin, CRP, MBL** Odense, Denmark: Allan Flyvbjerg

**Ultrasound reading centre** Pisa, Italy: M Kozakova

**ECG reading** Villejuif, France: MT Guillanneuf

**Actigraph** Villejuif, France: B Balkau, L Mhamdi

**Data Management** Villejuif, France, Padova, and Pisa, Italy: B Balkau, A Mari, L Mhamdi, L Landucci, S Hills, L Mota

**Mathematical modelling and website management** Padova, Italy: A Mari, G Pacini, C Cavaggion, A Tura

**Coordinating office:** Pisa, Italy: SA Hills, L Landucci, L Mota

Further information on the RISC Study and participating centres can be found on

www.egir.org
ONLINE SUPPLEMENT

For:

EUGLYCEMIC CLAMP INSULIN SENSITIVITY AND LONGITUDINAL SYSTOLIC BLOOD PRESSURE:

ROLE OF GENDER

John R Petrie, MD, PhD; Muhammad Omar Malik, MD; Beverley Balkau, PhD; Colin G Perry, MD, PhD; Kurt Højlund, MD; Zoltan Pataky, MD; John Nolan, MD; Ele Ferrannini, MD, PhD; Andrea Natali, MD, PhD

for the RISC Investigators

From the University of Glasgow, United Kingdom (J.R.P., M.O.M., C.P.); Khyber Medical University Peshawar, Pakistan (M.O.M); INSERM U1018, Villejuif, France (B.B.); University Paris Sud 11, UMRS 1018, Villejuif, France (B.B.); Odense University Hospital, Odense, Denmark (K.H.); University Hospital of Geneva, Switzerland (Z.P.); Steno Diabetes Center, Copenhagen, Denmark (J.N.); and the Department of Internal Medicine, University of Pisa, Pisa, Italy (E.F., A.N.)

Short title: Insulin sensitivity, gender and blood pressure

Corresponding Author:

Professor John R Petrie BSc, MBChB, PhD, FRCP(Ed), FRCPSG.
Professor of Diabetic Medicine
BHF Glasgow Cardiovascular Research Centre
University of Glasgow
126 University Place
Glasgow G12 8TA
Tel +44(0)141 330 3325
Fax +44(0)141 330 6972
E-mail john.petrie@glasgow.ac.uk
RISC investigators and recruiting centres:

**Amsterdam**, The Netherlands: RJ Heine, J Dekker, S de Rooij, G Nijpels, W Boorsma

**Athens**, Greece: A Mitrakou, S Tournis, K Kyriakopoulou, P Thomakos

**Belgrade**, Serbia: N Lalic, K Lalic, A Jotic, L Lukic, M Civcic

**Dublin**, Ireland: J Nolan, TP Yeow, M Murphy, C DeLong, G Neary, MP Colgan, M Hatunic

**Frankfurt**, Germany: T Konrad, H Böhles, S Fuellert, F Baer, H Zuchhold


Lehmann, T Merminod


**Kuopio**, Finland: M Laakso, U Salmenniemi, A Aura, R Raisanen, U Ruotsalainen, T

Sistonen, M Laitinen, H Saloranta

**London**, England: SW Coppack, N McIntosh, J Ross, L Pettersson, P Khadobaksh

**Lyon**, France: M Laville, F. Bonnet (now Rennes), A Brac de la Perriere, C Louche-Pelissier,

C Maitrepierre, J Peyrat, S Beltran, A Serusclat

**Madrid**, Spain: R. Gabriel, EM Sánchez, R. Carraro, A Friera, B. Novella


**Milan**, Italy: PM Piatti, LD Monti, E Setola, E Galluccio, F Minicucci, A Colleluori


Short, Y McGrady, D Richardson

**Odense**, Denmark: H Beck-Nielsen, P Staehr, K Hojlund, V Vestergaard, C Olsen, L Hansen

**Perugia**, Italy: GB Bolli, F Porcellati, C Fanelli, P Lucidi, F Calcinaro, A Saturni

**Pisa**, Italy: E Ferrannini, A Natali, E Muscelli, S Pinnola, M Kozakova, A Casolaro, BD

Astiarraga

**Rome**, Italy: G Mingrone, C Guidone, A Favuzzi, P Di Rocco
Vienna, Austria: C Anderwald, M Bischof, M Promintzer, M Krebs, M Mandl, A Hofer, A Luger, W Waldhäusl, M Roden

**Project Management Board:** B Balkau (Villejuif, France), SW Coppack (London, England), JM Dekker (Amsterdam, The Netherlands), E Ferrannini (Pisa, Italy), A Golay (Geneva, Switzerland), A Mari (Padova, Italy), A Natali (Pisa, Italy), JR Petrie (Glasgow, Scotland), M Walker (Newcastle, England)

**Core laboratories and reading centres:**

**Lipids** Dublin, Ireland: P Gaffney, J Nolan, G Boran

**Hormones** Odense, Denmark: C Olsen, L Hansen, H Beck-Nielsen

**Albumin:creatinine** Amsterdam, The Netherlands: A Kok, J Dekker

**Genetics** Newcastle-upon-Tyne, England: S Patel, M Walker

**Stable isotope laboratory** Pisa, Italy: A Gastaldelli, D Ciociaro

**Adiponectin, CRP , MBL** Odense, Denmark: Allan Flyvbjerg

**Ultrasound reading centre** Pisa, Italy: M Kozakova

**ECG reading** Villejuif, France: MT Guillanneuf

**Actigraph** Villejuif, France: B Balkau, L Mhamdi

**Data Management** Villejuif, France, Padova, and Pisa, Italy: B Balkau, A Mari, L Mhamdi, L Landucci, S Hills, L Mota

**Mathematical modelling and website management** Padova, Italy: A Mari, G Pacini, C Cavaggion, A Tura

**Coordinating office:** Pisa, Italy: SA Hills, L Landucci, L Mota

Further information on the RISC Study and participating centres can be found on

[www.egir.org](http://www.egir.org)