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ORIGINAL ARTICLE ***Reproductive endocrinology***

Insulin resistance in polycystic ovary syndrome: a systematic review and meta-analysis of euglycaemic–hyperinsulinaemic clamp studies

Samantha Cassar[1,2], Marie L. Misso[2], William G. Hopkins[1], Christopher S. Shaw[1,3], Helena J. Teede[2,4,†], and Nigel K. Stepto[1,2,*†]

[1]Institute of Sport Exercise and Active Living, College of Sport and Exercise Science, Victoria University, PO Box 14428, Melbourne, Victoria, 8001, Australia [2]Monash Centre for Health Research and Implementation, School of Public Health and Preventive Medicine, Monash University in Partnership with Monash Health, Locked Bag 29, Clayton, Victoria 3168, Australia [3]Institute for Physical Activity and Nutrition, School of Exercise and Nutrition Sciences, Deakin University, 75 Pigdons Road, Waurn Ponds, Victoria, 3126, Australia [4]Diabetes and Vascular Medicine Unit, Monash Health, 246 Clayton Road, Clayton, Victoria, 3168, Australia

*Correspondence address. Tel: +61-3-9919-5416; Fax: +61-3-9919-4891; Email: nigel.stepto@vu.edu.au

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STUDY QUESTION: What is the degree of intrinsic insulin resistance (IR) in women with polycystic ovary syndrome (PCOS) and the relative contribution of BMI to overall IR based on meta-analysis of gold standard insulin clamp studies?

SUMMARY ANSWER: We report an inherent reduction (−27%) of insulin sensitivity (IS) in PCOS patients, which was independent of BMI.

WHAT IS ALREADY KNOWN: PCOS is prevalent, complex and underpinned by IR but controversies surround the degree of intrinsic IR in PCOS, the effect of BMI and the impact of the different diagnostic criteria (NIH versus Rotterdam) in PCOS.

STUDY DESIGN, SIZE, DURATION: A systematic review and meta-analysis of Medline and All EBM databases was undertaken of studies published up to 30 May 2015. Studies were included if premenopausal women diagnosed with PCOS were compared with a control group for IS, measured by the gold standard euglycaemic–hyperinsulinaemic clamp. The systematic review adheres to the principles of the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) guidelines. Meta-analyses were performed using mixed modelling and magnitude-based inferences expressed as mean effect ±99% CI. We inferred the effect was small, moderate or large relative to a smallest important change of −3.7% or 3.8% derived by standardisation. Effects were deemed unclear when the CI overlapped smallest important positive and negative values. Effects were qualified with probabilities reflecting uncertainty in the magnitude of the true value (likely, 75–95%; very likely, 95–99.5%; most likely, >99.5%).

PARTICIPANTS/MATERIALS, SETTING, METHOD: A total of 4881 articles were returned from the search. Of these, 28 articles were included in the meta-analysis.

MAIN RESULTS AND THE ROLE OF CHANCE: Overall IS was lower in women with PCOS compared with controls (mean effect −27%, 99% CI ±6%; large, most likely lower). A higher BMI exacerbated the reduction in IS by −15% (±8%; moderate, most likely lower) in PCOS compared with control women. There was no clear difference in IS between women diagnosed by the original National Institutes of Health (NIH) criteria alone compared with those diagnosed by the Rotterdam criteria. Low levels of sex hormone-binding globulin (SHBG) were associated with reduced levels of IS (−10%, ±10%; small, very likely negative), which was not confounded by BMI.

LIMITATIONS, REASONS FOR CAUTION: This systematic review and meta-analysis inherited the confounding problems of small sample sizes, missing data (e.g. some hormones, waist and hip girths) and the lack of Rotterdam criteria phenotype reporting, limiting the evidence synthesis and meta-analysis.

[†]Joint senior authorship.

WIDER IMPLICATIONS OF THE FINDINGS: BMI has a greater impact on IS in PCOS than in controls. SHBG appears a potentially valuable marker of IR in PCOS, whereas testosterone after adjustment for BMI demonstrated an unexpected interplay with IS which warrants further investigation.

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REGISTRATION NUMBER: N/A

Key words: sex hormone-binding globulin / LH / FSH / testosterone / magnitude-based inferences

Introduction

Polycystic ovary syndrome (PCOS) is the most common endocrine and metabolic disorder affecting up to 18% of premenopausal reproductive aged women, depending on the population studied and diagnostic criteria used (March *et al.*, 2010). The condition has substantial short- and long-term reproductive, psychological and metabolic (insulin resistance [IR]) implications across the lifespan (Boomsma *et al.*, 2006; Teede *et al.*, 2011) and presents a significant health and economic burden. The estimated annual cost for the condition in the USA in 2004 was $4 billion, with 40% attributed to PCOS-associated diabetes (Azziz *et al.*, 2005). Hence, early diagnosis of PCOS and recognition of IR and metabolic complications are important to optimise screening, prevention and management.

The pathophysiology of PCOS is complex and remains elusive; however, IR (defined as reduced insulin sensitivity [IS] compared with control women) and hyperandrogenism play key roles in the aetiology (Teede *et al.*, 2007, 2010). It is generally accepted that obese women with PCOS are insulin resistant. Obesity is known to increase circulating androgen and insulin levels, may increase PCOS prevalence and exacerbates the clinical features of PCOS (Fig. 1; Diamanti-Kandarakis and Dunaif, 2012; Teede *et al.*, 2013). Consensus is yet to be achieved in lean women with PCOS as IR is not consistently demonstrated (Diamanti-Kandarakis and Dunaif, 2012). It is hypothesised that IR is intrinsic to PCOS and exacerbated by obesity (Dunaif and Graf, 1989; Stepto *et al.*, 2013), however, research is needed to establish whether IR in PCOS is independent of obesity.

Conflicting results are further confounded by evolving diagnostic criteria, with many studies using the original National Institutes of Health (NIH) Criteria, rather than the more inclusive Rotterdam criteria, now endorsed internationally (Diamanti-Kandarakis and Dunaif, 2012). IR may be more pronounced in the severe PCOS phenotype (original NIH criteria) of anovulation and hyperandrogenism, compared with those who have either normal androgen levels or regular menstrual cycles included under Rotterdam. All international guidelines now recommend Rotterdam criteria and recent genome-wide association studies in Caucasian (Day *et al.*, 2015; Hayes *et al.*, 2015) and Asian (Chen *et al.*, 2011; Shi *et al.*, 2012) PCOS populations report preservation of genes across both criteria. This suggests a more homogeneous disease and highlights the need for further research into features such as IR in women with Rotterdam diagnosed PCOS (Barber *et al.*, 2007; Panidis *et al.*, 2012).

IR and associated hyperinsulinaemia play a role in the reproductive, and endocrine features of PCOS by contributing to hyperandrogenism and disruption of gonadotrophin secretion (Norman *et al.*, 2007). IR and hyperinsulinaemia may directly stimulate production of androgens via ovarian tissue steroidogenesis, independently of changes in gonadotrophin concentration (Diamanti-Kandarakis and Dunaif, 2012). However, the relationships between IR in clamp studies and androgen levels require clarification. Hyperinsulinaemia also plays an indirect role in hyperandrogenism by inhibiting hepatic sex hormone-binding globulin (SHBG) production, thus increasing free testosterone (Kiddy *et al.*, 1989; Nestler *et al.*, 1991). Low SHBG levels are also increased by insulin-sensitising medication (Ehrmann *et al.*, 1997; Moghetti *et al.*, 2000; Moran *et al.*, 2013); however, the strength of the relationship between IR and SHBG is unclear and the potential role of SHBG as a clinical marker of IR in PCOS is yet to be determined.

IR can be measured using numerous direct and indirect techniques, including homeostatic model assessment, quantitative insulin sensitivity check index and oral glucose tolerance test-related measures comprising area under the curve for glucose and insulin (Muniyappa *et al.*, 2008). These are based on fasting or post glucose-load blood glucose and insulin levels, yet lack accuracy and reliability and are inadequate for clinical purposes and mechanistic research where IR is a primary study outcome (Muniyappa *et al.*, 2008). The euglycaemic–hyperinsulinaemic clamp is considered the reference standard to directly measure IS in research settings and can be used to define a specific cut-off level for IR (Muniyappa *et al.*, 2008). Yet, only limited studies on IR in PCOS have used euglycaemic–hyperinsulinaemic clamps.

In this common condition key knowledge gaps include the degree of IR in PCOS measured by gold standard clamp studies, the impact of BMI, the relationship to androgens, impact of diagnostic phenotypes and the role of SHBG as a clinically useful marker of IR. Therefore, we aimed to undertake a systematic review and meta-analysis to address the overarching question: What is the degree of intrinsic IR in PCOS and the relative contribution of BMI to overall IR based on meta-analysis of gold standard clamp studies? We also aimed to assess IR across different PCOS diagnostic criteria. Our secondary aim was to investigate key relationships between IR, BMI, SHBG and other reproductive hormones (testosterone, LH and FSH) in women with and without PCOS and assess the potential role of SHBG as a marker of IR.

Materials and Methods

We conducted a systematic review and a meta-analysis using mixed modelling and magnitude-based inferences on studies comparing IS measured by euglycaemic–hyperinsulinaemic clamp in lean and overweight women

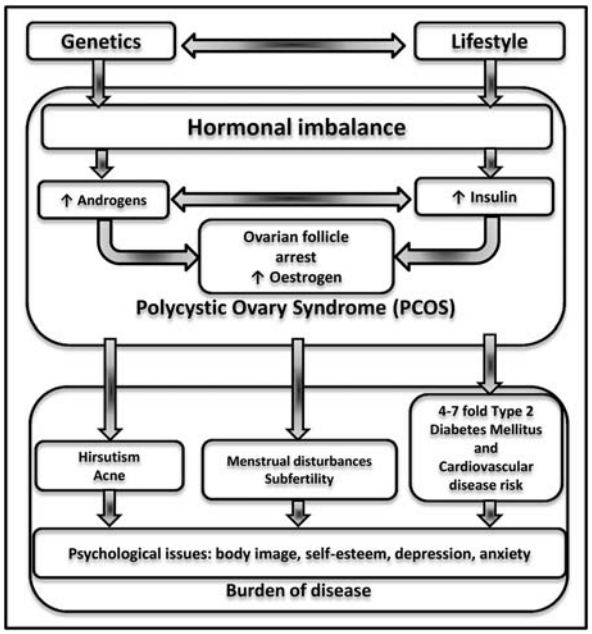

Figure 1 Diagnostic flow chart depicting the aetiological, hormonal and clinical features of polycystic ovary syndrome (PCOS). Adapted from Teede *et al.* (2011).

with and without PCOS. The methodology adheres to the Preferred Reporting Items for Systematic review and Meta-Analysis (PRISMA) Protocols. Authors are experienced in conducing systematic reviews and meta-analysis and work was directly undertaken by an experienced biostatistician.

Data sources and searches

A systematic search of published literature was conducted in Medline and All EBM databases (including Cochrane Database of Systematic Reviews, ACP Journal Club Database of Abstracts of Reviews of Effects, Cochrane Central Register of Controlled Trials, Cochrane Methodology Register, Health Technology Assessment and NHS Economic Evaluation Database) using the subject headings and key terms detailed in Table I. The search strategy was limited to English language articles. Bibliographies of included articles were also screened. The search was conducted on 30 September 2013 and updated on 30 May 2015.

Study selection

To determine the literature to be assessed further, two independent reviewers scanned the titles, abstracts and keywords of every record retrieved. Full texts of the remaining studies were retrieved for assessment according to the selection criteria determined *a priori*: PCOS was diagnosed by the NIH or the Rotterdam Criteria or equivalent (Fauser *et al.*, 2012) with the exclusion of other cause of hyperandrogenism; comparisons were made between at least one PCOS and control group; participants were premenopausal; 18–40 years of age; IS measured at baseline without any interventions, by a euglycaemic–hyperinsulinaemic clamp; control groups consisted of reproductive aged women 18–40 years who are reported as otherwise healthy and screened negative for features of PCOS based on the specified diagnostic criteria. Studies were excluded if participants: were taking medications or undergoing interventions that effect IS before baseline measures were assessed including insulin-sensitising medication, oral contraceptives, diet and exercise; were diagnosed with a confounding medical condition e.g. diabetes; or smoked. Disagreements were discussed and resolved by discussion and consensus with a third reviewer (N.S.).

Data extraction and quality assessment

Data were extracted from included studies using a specially developed data extraction form (Harris, 2010). Information was collected on study details, participants, results and validity of results. Corresponding authors were contacted by email if essential data were not reported in a usable format and if the study was published <10 years ago. Each email request to authors was sent using an institutional email account and provided a brief description of the systematic review scope, complete with article citation, specific information needed and our contact information. Authors were contacted a maximum of twice over a 4-week period. The study was not included in the meta-analysis if they failed to respond within this period. Authors of included papers were contacted for data according to PCOS phenotypes if the Rotterdam criteria were used. Data for IS and other subject characteristics were extracted and summarised as mean and SD; any SEs of the mean were converted to SD. To account for different units of IS measurement across studies, the statistical effect of PCOS on IS was expressed as a factor by dividing the IS in the PCOS group by the respective control group. We log-transformed all measures and used back-transformation to estimate meta-analysed overall relationship between IS and hormone effects as percentages with 99% CI (Snowling and Hopkins, 2006). Risk of bias of the included studies was assessed by a reviewer using criteria developed *a priori* (Harris, 2010). Findings from the body of evidence and their applicability to the research question were discussed in light of risk of bias. Disagreement or uncertainty was resolved by discussion to reach a consensus. Each study was then allocated a risk of bias rating (Supplementary Table S1).

Data synthesis and analysis

The log-transformed factor effects on IS were meta-analysed using the general linear mixed-model procedure (Proc Mixed) (Yang, 2003) in the Statistical Analysis System (Version 9.4, SAS Institute, Cary, NC, USA).

The first random-effects meta-analysis investigated the overall relationship between PCOS and IS. A random effect representing the identity of each study estimate was included to allow for true differences in the effect of PCOS between studies not accounted for by the other effects in the model. The weighting factor for each study estimate was the inverse of the square of the SE, derived as follows: the SEs of the mean in the PCOS and control groups were expressed as coefficients of variation (CV), converted to factors (1+CV/100), log-transformed, squared and added. The method of setting the residual variance to unity was used to apply the weighting (Yang, 2003).

The second meta-analysis explored the independent moderating effects of BMI, age and diagnostic criteria on the relationship between PCOS and IS. Fixed effects in the model were either linear numeric variables for BMI and age or a nominal variable for the diagnostic criteria (NIH or Rotterdam). The model with BMI as a predictor was used to compare predicted differences in IS in the PCOS and control groups at the weighted mean BMI of 22 and 32 kg/m^2, respectively, for lean and obese women (defined by a threshold BMI of 25 kg/m^2). The model for age as a predictor was used to compare differences in IS in the PCOS and control groups for younger and older women defined by the weighted mean of the means (27 years) and the weighted mean of the SDs (5 years) of the age of the women from each study. Hence, younger women were defined as aged 22 years (mean − 1 SD) and older as 32 years (mean + 1 SD).

Table I Search strategy for systematic review.[a]

1	exp[b] Polycystic Ovary Syndrome/
2	polycystic ovar$[c].mp[d].
3	poly-cystic ovar$.mp.
4	PCO$.mp.
5	(stein-leventhal or leventhal).mp.
6	Anovulation/
7	anovulat$.mp.
8	oligo-ovulat$.mp.
9	oligoovulat$.mp.
10	(ovar$ adj[e]5 (sclerocystic or polycystic or poly-cystic or degenerat$ or hyperandrogen$ or hyper-ndrogen$)).mp.
11	or/1–10
12	insulin resistan$.mp.
13	exp Insulin Resistance/
14	insulin resistance.mp.
15	insulin insensitiv$.mp.
16	insulin sensitiv$.mp.
17	exp Insulin/
18	insulin.mp.
19	exp Blood Glucose/
20	Blood Glucose.mp.
21	hyperinsulin$.mp.
22	glucose intolerance.mp.
23	euglycaemic-hyperinsulaemic clamp.mp.
24	euglycaemic hyperinsulaemic clamp.mp.
25	euglycemic-hyperinsulemic clamp.mp.
26	euglyc$ insulin clamp.mp.
27	insulin clamp.mp.
28	etiolog$.mp.
29	pathophysiol$.mp.
30	or/12–29
31	11 and 30
32	limit 31 to (English language and female and humans)

[a]Search was conducted for Medline with appropriate search terms utilised for other databases.
[b]exp = exploded.
[c]$ = any character.
[d]Medical Subject Heading for Medline: mp, title, abstract, original title, name of substance word, subject heading word, keyword heading word.
[e]adj = adjacency.

The third meta-analysis used a random-effects model in which fixed effects were used to explore the possible mutual confounding effects of BMI, age and diagnostic criteria on the difference in IS between PCOS and control groups. The effect of each moderator was therefore assessed while the other moderators were held constant.

The association between differences in key factors (testosterone, SHBG, LH and FSH) and differences in IS between PCOS and control groups were investigated in a fourth meta-analyses. In each analysis, the relationship between IR and PCOS was assessed using a random-effect model. A fixed-effect variable representing the difference in the log of the key factor between PCOS and control groups was then added to the model to determine the mediating effects of these key factors on IS. The difference in IS associated with each hormone was derived by multiplying the effect of the factor by the difference in the mean concentration of the factor between PCOS and control women. The possible confounding effect of BMI on the association between differences in key factors and differences in IS of PCOS and control groups was investigated by including BMI in the model.

Publication bias and outliers

We examined a scatter plot of the *t*-statistic associated with each study estimate value contributing to the study-estimate random effect versus the log of the SE of the effect. This plot is superior to the usual funnel plot (Hopkins *et al.*, 2009), because the *t*-value is effectively adjusted for uncertainty in the study estimates and for the contribution of study covariates. This approach identified three clear outliers (Micic *et al.*, 2007; Yang *et al.*, 2013; Wu *et al.*, 2014). Upon investigation of the data, it was noted that one study (Micic *et al.*, 2007) reported some measures as mean and SD instead of mean and SE as stated in Materials and Methods. After correction, this study was no longer an outlier. The other two studies were excluded from the meta-analysis (Supplementary Fig. S1).

Inferential statistics

Magnitudes of effects were evaluated via standardisation. For this purpose, a between-subject SD representing the typical variation in IS was derived from the square root of the weighted mean of the variances of the log-transformed factor SD for the healthy lean women in a subset of studies. The subset was determined by plotting the log of the factor SD against the mean BMI from healthy women in all studies. The plot showed a reasonably clear increase in the SD in studies with mean BMI >25 kg/m^2. We therefore used all the studies with a mean BMI of <25 kg/m^2 as the subset. Sample-size bias in the standardised effects was negligible, owing to the large number of degrees of freedom in the estimate of the SD and was therefore not corrected.

The effects in log-transformed units were divided by this SD, and their magnitudes were interpreted with the following scale: <0.20, trivial; 0.20–0.60, small; 0.60–1.2, moderate; >1.2, large (Hopkins *et al.*, 2009). In keeping with trends in inferential statistics (Sterne and Davey Smith, 2001; Snowling and Hopkins, 2006; Hopkins and Batterham, 2016), we made magnitude-based inferences about true population values of effects by expressing the uncertainty in the effects as 99% CIs. Effects were deemed unclear if the CIs overlapped thresholds for substantial positive and negative values (i.e. ±0.20 standardised units); effects were otherwise deemed clear and reported as the magnitude of its observed effect (Hopkins *et al.*, 2009). For an improvement in IS, magnitude thresholds were 3.8, 12, 25 and 46%, representing small, moderate, large and very large, respectively; for reduced IS, the corresponding magnitude thresholds were −3.7, −11, −20 and −31%, respectively. Magnitude-based inferences about effects were made more accurate and informative by qualifying the effects with qualitative probabilities that reflected the quantitative uncertainty in the magnitude of the true value (Hopkins *et al.*, 2009): possibly, 25–75%; likely, 75–95%; very likely, 95–99.5%; most likely, >99.5%.

Results

The original search returned 4369 articles (Fig. 2). Of these, 48 articles met the selection criteria but 23 of these were not included due to the following reasons: data not in a usable format and article published >10 years ago (eight articles); unable to obtain usable data from authors (four articles); data were already reported in a more current included article (11 articles). The remaining 25 articles were included in the final meta-analysis (Dunaif *et al.*, 1989; Ovesen *et al.*, 1993; Lasco *et al.*, 1995; Morin-Papunen *et al.*, 2000; Park *et al.*, 2001, 2007;

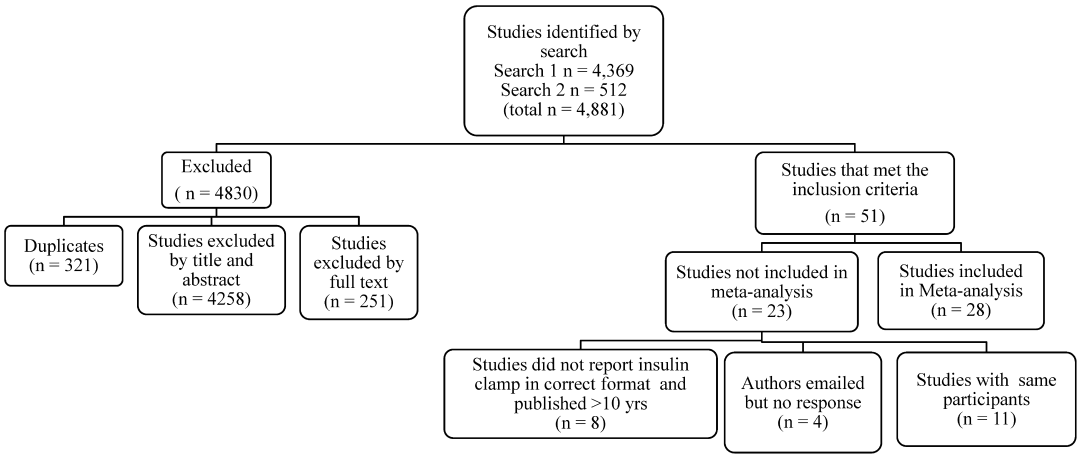

Figure 2 Search strategy results of included and excluded studies for a systematic review and meta-analysis of euglycaemic–hyperinsulinaemic clamp studies in PCOS.

Patel *et al.*, 2003; Vrbikova *et al.*, 2004; Baillargeon and Carpentier, 2007; Kowalska *et al.*, 2007, 2012; Micic *et al.*, 2007; Aroda *et al.*, 2008; Svendsen *et al.*, 2008; Ciaraldi *et al.*, 2009; Moret *et al.*, 2009; Oh *et al.*, 2009; Tosi *et al.*, 2009; Eriksen *et al.*, 2010, 2011; Nikolajuk *et al.*, 2010; Manneras-Holm *et al.*, 2011; Yang *et al.*, 2011; Li and Li, 2012; Stepto *et al.*, 2013). The updated search strategy resulted in 512 additional articles, of which, 32 were duplicate articles, 418 were excluded based on title and abstract, 58 were excluded based on full text or contact with authors (1 article) and 2 were excluded due to being identified as outliers, with potential publication bias on the funnel plot (Yang *et al.*, 2013; Wu *et al.*, 2014). The remaining three studies were included in the meta-analyses (Adamska *et al.*, 2013; Ciaraldi *et al.*, 2013; Leonhardt *et al.*, 2014), resulting in a total of 28 studies ($n = 741$ controls and $n = 1224$ PCOS) meeting the selection criteria. Some studies reported effects for more than one PCOS and control group within studies: these articles provided 38 study estimates. The effects from each study estimate are summarised in Table II. Rotterdam phenotype analysis on IR could not be performed owing to publications predating the criteria and/or insufficient responses to our requests for data from authors. A total of 24 studies were included in the meta-analysis investigating the moderating effects of key hormones (Table III).

A moderate risk of bias was reported in the majority of studies included in the systematic review and meta-analysis following quality appraisal (Supplementary Table S2).

Overall relationship between IS and PCOS

In the first simple meta-analysis model, IS was lower in women with PCOS compared with controls (a large difference; Table IV). Unexplained variance between studies was estimated as a CV of 21% (99% CI, ±10%).

Moderating effects of BMI on IS

Compared with their respective controls, lean and overweight women with PCOS showed lower IS with large and very large magnitudes respectively (Table IV, Fig. 3). Overweight women with PCOS had moderately lower IS compared with lean women with PCOS (Table IV) with weight contributing less to IR than to PCOS status. The residual between-study differences were 17% (±8%). Adjusting for differences in age and diagnostic criteria resulted in little change in the effect of BMI (Table IV) and little change in the residual between-study effect differences (18%, 99% CI, ±10%).

This BMI-only model was also able to produce separate effects of BMI on IS in PCOS and control women; a 10-unit higher BMI in women with PCOS was associated with a −28% (±14%; large, most likely) difference in IS. In control women, a 10-unit higher BMI resulted in a difference in IS of −15% (±17%; moderate, very likely lower).

Moderating effects of age on IS

Compared with their respective controls, younger and older women with PCOS showed lower or much lower IS, respectively (large and very large magnitudes). Older women diagnosed with PCOS had a lower IS compared with younger women with PCOS (a large magnitude), but this effect became small and unclear when BMI and diagnostic criteria were included in the model (Table IV). The residual between-study random-effect difference reported as a CV was 17% (±8%).

The second model was also able to estimate separate effects for age on IS in PCOS and control women. A 10-year increase in age in women with PCOS was associated with a −18% (±34%; unclear moderate effect) difference in IS; in control women the difference in IS for women 10 years older was 4% (±33%; unclear, small effect).

Moderating effects of diagnostic criteria as a surrogate for phenotype

The effect of PCOS on IS was large for both diagnostic criteria groupings and there was no difference between them (Table IV). The residual between-study differences in this analysis were a CV of 21% (±10%).

Table II Studies included in meta-analysis with age, BMI and insulin sensitivity in women with and without polycystic ovary syndrome (PCOS) and the effect of PCOS on insulin sensitivity.

Study	Sample size (Control, PCOS)	Diagnostic criteria	Age (years) Mean ± SD		BMI (kg/m^2) Mean ± SD		Insulin sensitivity[a] Mean ± SD		Effect of PCOS on insulin sensitivity[b] (%) Mean (90% CI)
			Control	PCOS	Control	PCOS	Control	PCOS	
Aroda et al. (2008)	6, 31	NIH	32 ± 5	29 ± 6	36.2 ± 6.9	35.3 ± 7.4	8.8 ± 2.0	5.6 ± 2.9	−36.2 (−48.7 to −20.7)
Adamska *et al.* (2013)	14, 40	Rotterdam	27 ± 6	24 ± 4	22.0 ± 2.1	21.4 ± 2.0	12.4 ± 2.9	10.0 ± 2.9	−19.4 (−29.1 to −8.3)
Adamska *et al.* (2013)	16, 54	Rotterdam	27 ± 7	26 ± 7	31.2 ± 4.8	31.2 ± 4.1	8.7 ± 4.2	7.2 ± 3.3	−17.2 (−33.7 to 3.3)
Baillargeon and Carpentier (2007)	17, 9	NIH	31 ± 7	24 ± 7	22.0 ± 2.1	22.6 ± 1.2	52.9 ± 19.0	48.5 ± 18.9	−8.3 (−29.2 to 18.8)
Ciaraldi *et al.* (2009)	15, 42	NIH	32 ± 4	29 ± 7	33.9 ± 7	35.4 ± 7.1	9.7 ± 2.8	6.5 ± 2.2	−32.8 (−42.2 to 21.9)
Ciaraldi *et al.* (2013)	12, 20	NIH	32 ± 1	28 ± 1	39.4 ± 2.2	35.6 ± 1.5	8.6 ± 0.7	5.8 ± 0.4	−32.6 (−43.3 to −20.0)
Dunaif *et al.* (1989)	8, 10	NIH	29 ± 6	27 ± 6	21.3 ± 1.1	22.3 ± 1.6	7.4 ± 1.1	4.8 ± 1.6	−35.9 (−47.8 to −21.6)
Dunaif *et al.* (1989)	11, 17	NIH	30 ± 3	26 ± 5	33.3 ± 5.6	34.3 ± 4.6	3.8 ± 1.5	2.4 ± 0.7	−36.0 (−49.0 to −19.7)
Eriksen *et al.* (2010)	14, 28	NIH	33 ± 8	31 ± 6	33.7 ± 6.4	33.2 ± 4.2	297 ± 86.1	150 ± 42.3	−49.5 (−56.8 to −41.0)
Eriksen *et al.* (2011)	8, 8	NIH	32 ± 10	33 ± 3	35.1 ± 5.9	34.5 ± 4.0	271 ± 78.3	147 ± 23.1	−45.9 (−55.7 to −34.0)
Kowalska *et al.* (2007)	25, 23	Rotterdam	26 ± 6	24 ± 4	21.8 ± 2.0	21.4 ± 2.1	11.6 ± 3.0	9.1 ± 3.5	−21.4 (−32.8 to −8.1)
Kowalska *et al.* (2007)	20, 47	Rotterdam	28 ± 7	26 ± 6	31.0 ± 4.4	31.0 ± 4.0	9.4 ± 3.1	7.3 ± 3.1	−22.6 (−34.0 to −9.3)
Kowalska *et al.* (2012)	28, 65	Rotterdam	27 ± 6	25 ± 6	28.2 ± 6.8	27.3 ± 7.2	58.1 ± 22.3	45.7 ± 18.7	−21.2 (−31.8 to −9.1)
Lasco *et al.* (1995)	6, 10	Rotterdam[c]	28 ± 2	24 ± 2	21.0 ± 0.8	36.4 ± 2.2	5.8 ± 2.4	2.8 ± 1.1	−51.7 (−66.1 to −31.3)
Leonhardt *et al.* (2014)	31, 58	Rotterdam	28 ± 3.5	30 ± 4.5	23.6 ± 4.9	24.9 ± 4.8	13 ± 4.1	11 ± 3	−15.4 (−24.2 to −5.6)
Li and Li (2012)	92, 78	NIH	26 ± 3	25 ± 5	20.5 ± 1.6	20.7 ± 1.8	12.4 ± 1.7	10.1 ± 2.5	−18.6 (−22.8 to −14.3)
Li and Li (2012)	92, 33	NIH	26 ± 3	25 ± 5	20.5 ± 1.6	26.8 ± 2.4	12.4 ± 1.7	7.46 ± 1.8	−39.8 (−44.2 to −35.2)
Manneras-Holm *et al.* (2011)	31, 31	Rotterdam	28 ± 4	29 ± 3	24.7 ± 4.9	24.8 ± 4.8	13.0 ± 4.1	11.0 ± 3.0	−15.38 (−25.1 to −4.41)
Micic *et al.* (2007)	8, 8	Rotterdam	25 ± 20	22 ± 8	20.2 ± 3.3	20.5 ± 3.7	7.8 ± 3.7	4.4 ± 2.2	−43.1 (−61.5 to −16.0)
Micic *et al.* (2007)	8, 8	Rotterdam	29 ± 14	25 ± 18	31.0 ± 10.4	34.4 ± 18.5	3.92 ± 2.9	1.82 ± 1.8	−53.6 (−76.3 to −9.2)
Moret *et al.* (2009)	5, 5	Rotterdam	21 ± 2	24 ± 4	21.3 ± 1.0	23.0 ± 4.3	9.8 ± 2.0	8.2 ± 2.7	−16.3 (−38.6 to 14.0)
Morin-Papunen *et al.* (2000)	17, 15	Rotterdam	37 ± 3	29 ± 5	22.9 ± 1.2	22.7 ± 1.9	48.2 ± 9.9	41.1 ± 14.3	−14.7 (−28.1 to 1.1)
Morin-Papunen *et al.* (2000)	17, 28	Rotterdam	35 ± 5	30 ± 5	31.8 ± 4.7	34.5 ± 5.3	31.6 ± 11.1	20.5 ± 7.9	−35.1 (−46.0 to −22.1)
Nikolajuk *et al.* (2010)	18, 35	Rotterdam	26 ± 6	24 ± 4	22.2 ± 1.9	21.7 ± 1.8	8.9 ± 2.3	7.2 ± 2.9	−18.5 (−29.7 to −5.7)
Nikolajuk *et al.* (2010)	16, 43	Rotterdam	27 ± 5	26 ± 6	30.7 ± 4.4	31.5 ± 4.3	5.9 ± 2.2	4.5 ± 2.4	−24.4 (−38.0 to −7.8)
Oh *et al.* (2009)	24, 39	NIH	26 ± 1	25 ± 1	19.9 ± 0.3	20.8 ± 0.2	6.3 ± 0.3	5.3 ± 1.4	−15.9 (−21.7 to −9.6)
Ovesen *et al.* (1993)	7, 7	NIH	26 ± 4	21 ± 5	21.3 ± 1.8	22.2 ± 2.1	3.8 ± 1.3	4.0 ± 1.1	5.3 (−20.4 to 39.2)
Park *et al.* (2001)	5, 9	NIH	31 ± 11	25 ± 12	25.6 ± 5.3	26.0 ± 9.3	9.4 ± 5.1	2.3 ± 0.9	−75.5 (−84.9 to −60.5)
Park *et al.* (2007)	34, 73	NIH	26 ± 3	25 ± 4	20.9 ± 3.2	20.4 ± 1.5	6.7 ± 1.6	5.3 ± 1.3	−20.9 (−27.0 to −14.3)
Patel *et al.* (2003)	9, 11	NIH	26 ± 2	29 ± 2	27.4 ± 2.1	35.3 ± 2.7	8.3 ± 2.4	5.2 ± 3.3	−37.4 (−55.8 to −11.3)
Stepto *et al.* (2013)	19, 20	Rotterdam	28 ± 6	27 ± 4	22.0 ± 2.0	23.0 ± 2.0	339 ± 76	269 ± 66	−20.7 (−29.8 to −10.2)
Stepto *et al.* (2013)	14, 20	Rotterdam	35 ± 4	30 ± 6	35.0 ± 6	36.0 ± 7.0	264 ± 66	175 ± 96	−33.7 (−47.1 to −17.0)
Svendsen *et al.* (2008)	9, 17	Rotterdam	20 ± 4	28 ± 5	22.0 ± 1.4	23.0 ± 1.5	13.3 ± 4.1	10.4 ± 3	−21.8 (−32.4 to −9.6)
Svendsen *et al.* (2008)	16, 18	Rotterdam	31 ± 5	29 ± 4	34.0 ± 3.2	33.0 ± 4.0	8.1 ± 2.8	6.9 ± 2.0	−14.8 (−28.9 to 2.0)

Tosi *et al.* (2009)	35, 50	NIH	25 ± 5	22 ± 4	23.4 ± 5	24.0 ± 4	13.9 ± 2.0	10.3 ± 2.8	−25.9 (−31.2 to −20.2)
Vrbikova *et al.* (2004)	15, 53	NIH	28 ± 6	24 ± 5	21.5 ± 2.0	21.5 ± 1.8	43.9 ± 11.1	41.7 ± 12.2	−4.9 (−16.3 to 8.1)
Vrbikova *et al.* (2004)	15,[d] 30	NIH	28 ± 6	26 ± 5	21.5 ± 2.0	29.6 ± 3.7	43.9 ± 11.1	32.2 ± 10.0	−26.5 (−36.3 to −15.3)
Yang *et al.* (2011)	116, 133	Rotterdam	26 ± 3	25 ± 4	21.0 ± 2.2	23.5 ± 3.5	12.1 ± 1.8	8.4 ± 2.8	−30.6 (−34.2 to −26.9)
Overall weighted means			28 ± 5	26 ± 5	24.7 ± 3.9	26.4 ± 4.4	ANA[a]	ANA[a]	−26.7 (−30.1 to −23.1)[e]

NIH, National Institutes of Health; PCOS, polycystic ovary syndrome. The effects of PCOS on insulin sensitivity from each study are factor effects presented as percentages with 90% CI calculated from means, SD and sample size in PCOS and control groups.
[a]Units of measure for insulin sensitivity not included as units and calculations differed between studies and cannot obtain weighted means from raw data (ANA; analysis not available).
[b]Difference in insulin sensitivity between PCOS and control women as a percentage of control.
[c]Diagnostic criteria—defined as Rotterdam criteria based on diagnosis described in paper despite predating the published consensus Fauser *et al.* (2012).
[d]Overweight control participants were not reported. Therefore, comparisons were made between the lean control and overweight PCOS group.
[e]Meta-analysed effect (no-covariates).

IS, SHBG and hormone concentrations

Table V shows the uncorrected and BMI-corrected associations of the differences in concentrations of SHBG, testosterone, LH and FSH with differences in IS, comparing PCOS and control women. The mean lower concentration of SHBG in PCOS compared with controls was associated with a small lower IS, which was not confounded by BMI. Testosterone means were higher in PCOS and were associated with worse IS but the relationship indicates women with less testosterone are more insulin resistant at the same BMI. This implies that if there was no difference in testosterone between PCOS and control participants, women with PCOS would have even lower IS (−29%, ±10% and 41%, ±15% (large, most likely) for BMI's of 22 and 32 kg/m^2, respectively). Higher LH concentrations in women with PCOS tended (unclear effect) to be associated with lower IS, while FSH had a trivial relationship with only minor differences in FSH concentration between PCOS and control women.

Discussion

In this novel and comprehensive systematic review and meta-analysis, we report that women with PCOS have 27% worse IS as a measure of IR from the gold standard euglycaemic–hyperinsulinaemic clamp, than controls, independent of BMI, age or diagnostic criteria. BMI independently exacerbated IR by 15% in PCOS, and had a greater impact than in controls. Increasing age adversely impacted IR, yet the impact was trivial when adjusted for BMI and diagnostic criteria. PCOS diagnostic criteria had a trivial effect on IR. SHBG had a strong negative association with IR, which was not confounded by BMI. Total testosterone, when the confounding effects of BMI were controlled, had a moderate and very likely effect on IR. The relationship of LH with IR was moderate and unclear when BMI was included in the model and FSH concentrations did not differ between PCOS and controls.

A key role of insulin is to regulate glucose homeostasis by stimulating glucose uptake in target tissues including skeletal muscle and adipocytes and by suppressing hepatic glucose production (Cho *et al.*, 2011). IR can be defined as an impairment of insulin to mediate metabolism in skeletal muscle, adipocytes and liver. This includes glucose uptake, glycogen synthesis and inhibition of lipolysis, resulting in compensatory hyperinsulinaemia to achieve glucose homeostasis (Kahn and Flier, 2000; Diamanti-Kandarakis and Dunaif, 2012). Measurement of IR in clinical practice is not recommended as methods remain inaccurate (Teede *et al.*, 2011). However in research, the gold standard for assessing IR *in vivo* is generally accepted to be the euglycaemic–hyperinsulinaemic clamp (DeFronzo *et al.*, 1979). Using this technique, PCOS has an intrinsic IR (Dunaif *et al.*, 1989; Stepto *et al.*, 2013), however, data are inconsistent, especially in lean women with PCOS and the degree of IR intrinsic to PCOS has not been defined. Limitations have included small sample sizes and not accounting for variations in ethnicity, or diagnostic categories. Here, we address these gaps and advance the field by confirming in a robust systematic review and meta-analysis, that PCOS is underpinned by an intrinsic IR (~27% reduction in IS), independent of BMI and age, as well as across different diagnostic criteria and ethnic groups.

We further progress understanding in this area by showing that increased BMI exacerbates IR by 15% and has a greater adverse impact on IR in PCOS than in controls. This may be related to potentially

Table III Studies included in the meta-analysis investigating moderating effects of key hormones.

Study	Sex hormone-binding globulin (nmol/L) Mean ± SD		Testosterone (nmol/L) Mean ± SD		LH (IU/L) Mean ± SD		FSH (IU/L) Mean ± SD	
	Control	PCOS	Control	PCOS	Control	PCOS	Control	PCOS
Aroda et al. (2008)	22 ± 11.8	20 ± 15.6	0.7 ± 0.3	1.3 ± 4.9				
Adamska et al. (2013)	87.5 ± 56.1	59.6 ± 42.2	1.9 ± 0.3	2.8 ± 1.3	5.2 ± 2.1	9.8 ± 6.0	6.0 ± 1.4	5.8 ± 1.7
Adamska et al. (2013)	36.7 ± 18.4	43.6 ± 35.5	1.7 ± 0.7	2.7 ± 7.6	4.7 ± 2.2	7.9 ± 3.6	5.2 ± 1.8	5.4 ± 1.7
Baillargeon and Carpentier (2007)							4.2 ± 1.2	4.7 ± 1.5
Ciaraldi et al. (2009)	17.6 ± 13.9	17.5 ± 12.3	0.9 ± 0.5	1.9 ± 1.1				
Ciaraldi et al. (2013)	23.8 ± 20.1	16.6 ± 8.5	2.4 ± 1.2	5.0 ± 2.5				
Dunaif et al. (1989)	26.7 ± 10.8	28.5 ± 15.4	1.0 ± 0.4	1.6 ± 0.6				
Dunaif et al. (1989)	20.1 ± 8.7	15.6 ± 4.5	0.7 ± 0.2	1.8 ± 1.1				
Kowalska et al. (2007)	87.1 ± 47.5	69.3 ± 38.1	1.8 ± 0.5	2.7 ± 0.9	6.6 ± 4.5	11.7 ± 6.2	6.3 ± 1.5	5.9 ± 1.3
Kowalska et al. (2007)	47.6 ± 28.7	41.8 ± 33.6	1.8 ± 0.3	2.8 ± 1.2	4.7 ± 2.2	8.2 ± 3.6	5.3 ± 1.9	5.6 ± 1.4
Kowalska et al. (2012)	59.8 ± 48.8	41.5 ± 22.0	1.4 ± 0.3	2.1 ± 0.9	4.7 ± 2.1	7.9 ± 4.9	5.5 ± 1.6	6.0 ± 1.4
Lasco et al. (1995)			2.9 ± 1.1	1.6 ± 0.7	7.9 ± 2.3	3.9 ± 1.1	4.8 ± 0.9	5.4 ± 1.1
Li and Li (2012)			1.6 ± 0.6	1.9 ± 0.7				
Li and Li (2012)			1.6 ± 0.6	2.1 ± 0.9				
Manneras-Holm et al. (2011)	69.4 ± 30.4	49.0 ± 25.1	0.7 ± 0.3	1.5 ± 0.7				
Micic et al. (2007)	54.7 ± 48.9	26.2 ± 24.5	1.9 ± 2.0	3.6 ± 6.7				
Micic et al. (2007)	43.2 ± 40.6	15.8 ± 20.0	2.1 ± 1.5	4.9 ± 8.6				
Moret et al. (2009)			1.2 ± 0.1	1.8 ± 0.8	3.8 ± 1.8	13.6 ± 3.4	5.2 ± 0.8	6.1 ± 1.5
Morin-Papunen et al. (2000)	60.5 ± 23.5	43.0 ± 16.7	1.3 ± 0.8	2.0 ± 0.8	4.7 ± 1.6	7.5 ± 2.7	6.2 ± 1.6	5.1 ± 1.9
Morin-Papunen et al. (2000)	51.0 ± 27.6	30.8 ± 12.7	1.3 ± 0.4	2.3 ± 1.1	5.3 ± 3.3	7.0 ± 2.6	7.9 ± 5.8	6.4 ± 2.6
Nikolajuk et al. (2010)	89.5 ± 56.2	59.9 ± 46.7	1.8 ± 0.4	2.9 ± 1.1	6.9 ± 5.6	10.2 ± 5.3	6.0 ± 1.3	5.8 ± 1.5
Nikolajuk et al. (2010)	41.5 ± 18.4	36.2 ± 18.0	1.8 ± 0.5	2.8 ± 1.1	4.3 ± 2.2	9.0 ± 4.0	5.6 ± 1.7	5.5 ± 1.4
Oh et al. (2009)	56 .0 ± 7.0	43.0 ± 4.0						
Ovesen et al. (1993)	79.3 ± 12.2	59.9 ± 13.8	1.0 ± 0.3	2.9 ± 0.9				
Park et al. (2001)	89.1 ± 51.7	20.1 ± 37.5	1.1 ± 1.0	0.9 ± 0.5	12.0 ± 7.2	22.3 ± 16.6	9.0 ± 5.4	10.0 ± 9.6
Park et al. (2007)	54.7 ± 25.7	50.5 ± 23.9			4.4 ± 4.3	10.2 ± 5.6	4.1 ± 2.2	5.5 ± 1.7
Patel et al. (2003)			1.2 ± 0.3	2.6 ± 0.3	3.4 ± 1.5	7.9 ± 5.0	4.8 ± 1.5	4.0 ± 1.3
Stepto et al. (2013)	79.0 ± 19.0	69.0 ± 34.0	1.5 ± 0.5	2.1 ± 0.8				
Stepto et al. (2013)	46.0 ± 29.0	32.0 ± 11.0	1.5 ± 0.8	2.6 ± 0.8				
Svendsen et al. (2008)	104 ± 33.0	67.0 ± 27.0	1.5 ± 0.3	2.1 ± 0.8				
Svendsen et al. (2008)	54.0 ± 21.0	57.0 ± 39.0	1.4 ± 0.4	2.4 ± 0.8				
Tosi et al. (2009)	72.0 ± 7.0	63.0 ± 6.0	2.1 ± 1.0	3.1 ± 1.5				
Vrbikova et al. (2004)	68.5 ± 21.3	48.6 ± 24.3	1.8 ± 0.6	3.4 ± 1.3	5.4 ± 2.1	7.6 ± 5.2	4.8 ± 1.8	
Vrbikova et al. (2004)	68.5 ± 21.3	36.0 ± 22.7	1.8 ± 0.6	3.3 ± 1.5	5.4 ± 2.1	5.9 ± 2.9	4.8 ± 1.8	
Yang et al. (2011)			1.5 ± 0.7	2.0 ± 1.0				
Overall mean ± SD	57.0 ± 23.9	40.7 ± 17.2	1.4 ± 0.4	2.4 ± 0.8	5.3 ± 2.0	9.6 ± 3.9	5.8 ± 1.3	5.7 ± 1.3

greater visceral fat in PCOS, at least in the overweight and obese group (Lim et al., 2012). Further research is needed to determine the mechanisms by which BMI disproportionately increases IR in PCOS and to identify effective interventions to prevent and manage obesity in PCOS (Teede et al., 2011). Overall, given that women with PCOS have increased IR, higher BMI, and higher rates of weight gain that drive clinical severity (Teede et al., 2013), maintenance of healthy weight through lifestyle and exercise is a vital clinical focus. Lifestyle modification, which improves IR, is the first-line treatment in PCOS (Teede et al., 2013), including aerobic exercise with efficacy even in the absence of weight loss (Hutchison et al., 2011). Effective, safe insulin sensitisers are needed for PCOS. Our recent systematic review and meta-analysis shows metformin, an insulin sensitiser, in addition to lifestyle modification induces greater loss of body fat and improves symptoms compared with lifestyle alone (Teede et al., 2007; Misso and Teede, 2015; Naderpoor et al., 2015).

Table IV Meta-analysed overall mean effect of PCOS and insulin sensitivity, the predicted unadjusted and adjusted effects in study subgroups defined by BMI, age and diagnosis, and the effects of each of these moderators.

	Unadjusted effects		Adjusted effects	
	Effect, ±CI (%)	Qualitative inference	Effect, ±CI (%)	Qualitative inference
Overall mean	−27, ±6	Large****		
Moderation by mean BMI				
BMI=22 kg/m^2 (lean)	−20, ±6	Moderate****	−20, ±8[†]	Large****
BMI=32 kg/m^2 (overweight)	−31, ±6	Very large****	−30, ±9[†]	Very large****
Overweight PCOS versus lean PCOS	−15, ±9	Moderate****	−13, ±13[†]	Moderate***
Moderation by mean age				
Age=22 years (younger)	−17, ±14	Moderate***	−23, ±15[‡]	Large****
Age=32 years (older)	−35, ±12	Very large****	−28, ±13[‡]	Large****
Older PCOS versus younger PCOS	−21, ±23	Large (unclear)	−7, ±32[‡]	Small (unclear)
Moderation by diagnosis				
Rotterdam	−24, ±8	Large****	−23, ±8[§]	Large****
NIH	−30, ±8	Large****	−28, ±8[§]	Large****
NIH versus Rotterdam	−8, ±14	Small**	−6, ±11[§]	Small (unclear)

Mean ± 99% confidence limits.
Superscript alphabets (*, **, ***, ****) indicate likelihood that the true effect is substantial. A substantial reduction is reported as follows: *possibly, **likely, ***very likely, ****most likely.
[†]Adjusted to mean age (27 years) and mean diagnosis ((Rotterdam+NIH)/2).
[‡]Adjusted to mean BMI (27 kg/m^2) and mean diagnosis.
[§]Adjusted to mean age and mean BMI.

The recent NIH consensus workshop on PCOS endorsed the Rotterdam criteria as the definitive diagnostic criteria (National Institutes of Health, 2012) and recommended that all studies include analyses by reproductive PCOS phenotype. However, little prior PCOS literature has reported by phenotype, necessitating analysis here by diagnostic criteria as a proxy. We report that woman with original NIH-diagnosed PCOS alone (irregular cycles and hyperandrogenism) were 30% more insulin resistant compared with controls. Prior studies suggest that women with an NIH diagnosis of PCOS are more IR than those diagnosed via Rotterdam criteria. However, most studies have used inaccurate measures of IR (Dewailly *et al.*, 2006; Barber *et al.*, 2007; Panidis *et al.*, 2012). In our current work, we advance the field by showing that women diagnosed under the broader Rotterdam criteria (two criteria of either: irregular cycles, polycystic ovaries and/or hyperandrogenism) are 23% more insulin resistant than controls. We also show similar rates of IR in studies using original NIH versus those using Rotterdam criteria. Similarities in IR may be related to genetic variations that are preserved across these criteria (Chen *et al.*, 2011; Day *et al.*, 2015; Hayes *et al.*, 2015), suggesting a homogenous disease origin linked to insulin-related genes (Shi *et al.*, 2012). In most studies women with a 'Rotterdam' diagnosis of PCOS include those meeting NIH criteria, a limitation of this review, although we have previously shown that even young lean women diagnosed by Rotterdam criteria alone are insulin resistant (Stepto *et al.*, 2013). This and the lack of reporting by PCOS phenotype highlight the need for further research.

Our meta-analysis allowed exploration of potential mediators or moderators of IR in PCOS, including SHBG, testosterone, LH and FSH. Here, we report that in PCOS compared with controls SHBG had a strong inverse association with IR, confirming previous studies (Wallace *et al.*, 2013). This is consistent with mechanistic studies demonstrating that SHBG production is suppressed by IR and hyperinsulinaemia (Le *et al.*, 2012) and that SHBG concentrations increase with insulin sensitiser treatment (Moghetti *et al.*, 2000). Our work supports the use of SHBG as a simple clinical marker of IR in women with PCOS. We also confirm that SHBG is not only lower, but also less variable in PCOS, compared with controls (Jayagopal *et al.*, 2003), with larger studies now needed to derive specific SHBG cut-off ranges to predict IR. Furthermore, low SHBG is associated with adverse cardiovascular risk factors, diabetes and metabolic syndrome, independent of obesity, all important clinical implications of IR in PCOS, suggesting that SHBG may also be a useful clinical marker of metabolic risk in PCOS.

The most consistently observed androgenic abnormality in PCOS is elevated 'free testosterone' or calculated free androgen index, which includes SHBG in the equation (Teede *et al.*, 2011). This introduces a confounder, and given that IR has a profound effect on SHBG, relationships between androgens and IR should be studied independent of SHBG. Here, we focussed on studies that reported total testosterone levels. Our meta-analysis suggested a very likely moderate inverse relationship, with higher androgens associated with reduced IR, once corrected for BMI. This is in contrast to our current understanding of the interplay between androgens and IR (Diamanti-Kandarakis and Dunaif, 2012). However, caution is needed when interpreting these data as radioimmunoassays (n = 10) and chemiluminescence immunoassays (n = 9) were predominately used for the measurement of androgens, which are less sensitive than mass spectrometry (n = 2) in detecting androgen levels in women (Handelsman and Wartofsky, 2013). As we transition to more accurate methods, greater insights may be gained into the interrelationships between hyperandrogenism and IR in PCOS.

The unexplained residual differences between studies, represented by the random effect, were large (~21%) but fell to moderate, when all predictors were included in the model (Hopkins et al., 2009). Therefore, the overall meta-analytic model did not account for all the between-study variation in the effect of PCOS on insulin resistance. Differences in study or subject characteristics, including inherent problems in analysing and reporting data from the euglycaemic–hyperinsulinaemic clamp, recruitment strategies, ethnicity and lifestyle factors (Day et al., 2015) may account for some of the unexplained effects of PCOS on IR. Other key factors may also influence IR in PCOS, including low-grade inflammation (Shorakae et al., 2015) and sympathetic nervous system dysfunction (Diamanti-Kandarakis and Dunaif, 2012; Shorakae et al., 2015) and further research is needed to clarify key mechanisms underpinning IR in PCOS.

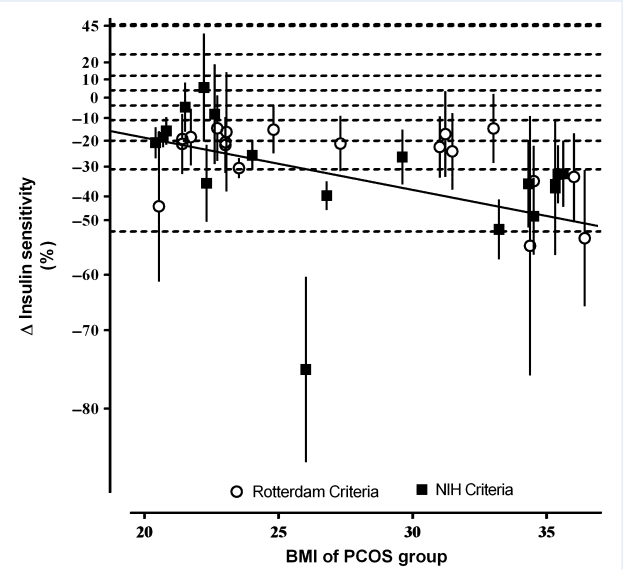

Figure 3 The effect of BMI on the difference in insulin sensitivity between PCOS and control groups. The dotted lines represent magnitude-based thresholds. For an improvement in insulin sensitivity, magnitude thresholds were 3.8, 12, 25, 48 and 120%, representing small, moderate, large, very large and extra large, respectively; for reduced insulin sensitivity, the corresponding magnitude thresholds were −3.7, −11, −20, −31 and −53%.

The strength of this meta-analysis is the extensive and comprehensive literature search and the focus on studies using gold standard methods to measure IR. However, as with many systematic reviews and meta-analyses, there are limitations. A meta-analysis cannot solve inherent confounding problems in included studies, which may exaggerate or underestimate effects. There was a lack of available robust trials and challenges of extracting reported data from the included studies as outlined. None of the studies included in the meta-analysis reported if the person running the clamps was blinded to the diagnosis of the participant. The majority of articles included in this analysis were of moderate to high risk of bias. Furthermore, not all identified studies were included because of difficulties sourcing the required data from original authors. The inability to acquire missing data from all eligible studies is not unexpected and deemed part of the meta-analysis process (Kelley et al., 2004). Inconsistent reporting of results made it necessary to express variables as factors by dividing the given variable in the PCOS group by that in the respective control group. Also endpoints varied (including units and scales). The body of evidence was heterogeneous such that diagnostic criteria were not uniformly applied with many studies only including women with NIH-diagnosed PCOS, and few reporting reproductive phenotypes. Small sample sizes and missing data limited evidence synthesis and meta-analysis. Despite these limitations, this body of work considerably advances the field by demonstrating that women with PCOS have an intrinsic IR.

Conclusion

We have completed the first comprehensive systematic review and meta-analysis of IR on euglycaemic–hyperinsulinaemic clamp studies in women with PCOS compared with controls. We have included 28

Table V Associations of differences in hormone concentrations with the difference in insulin sensitivity between women with and without PCOS.

Hormones	Percent difference in [Hormone][a] Mean ± SD	Unadjusted effects		Adjusted effects	
		Effect[b] (%) Mean, ±99%CI	Qualitative inference	Effect[c] (%) Mean, ±99%CI	Qualitative inference
Sex hormone-binding globulin (*n*=16)	−25 ± 21	−10, ±10	Small ↓***	−10, ±10	Small ↓**
LH (*n*=10)	88 ± 56	−3, ±44	Trivial (unclear)	−11, ±27	Moderate (unclear)
Testosterone (*n*=20)	76 ± 42	7, ±21	Small (unclear)	18, ±18	Moderate ↑***
FSH (*n*=10)	0.9 ± 14	0, ±0.1	Trivial****	0, ±0.1	Trivial****

Superscript alphabets (*, **, ***, ****) indicate the likelihood that the true effect is substantial. A substantial reduction is reported as follows: *possibly, **likely, ***very likely, ****most likely.
↓ Indicates lower insulin sensitivity,
↑ indicates higher insulin sensitivity
n indicates the number of studies included.
[a]Indicates the between-study means and SD of the difference in the mean concentration of hormones in PCOS and control women.
[b]Indicates the difference in insulin sensitivity associated with the mean of the difference in the means (PCOS − control) of the hormone concentrations.
[c]Indicates the difference in insulin sensitivity associated with the mean of the difference in the means (PCOS − control) of the hormone concentrations when BMI is taken into account.

studies (741 women with PCOS and 1224 controls) with an overall moderate risk of bias. We report that PCOS has an intrinsic IR with a 27% lower IS, independent of BMI, with results that are statistically qualitatively 'large' and 'most likely', as defined here. We also report a novel finding that BMI not only independently exacerbates IR in PCOS but has a disproportionately greater impact on IR in PCOS than in controls. We demonstrate that expanding diagnostic criteria to include the more inclusive Rotterdam criteria (encompassing original NIH criteria) appears to have a limited impact on IR in PCOS. Future studies need to focus on IR across the PCOS phenotypes, as most studies fail to report phenotype. This work supports the critical need for lifestyle management, including exercise and dietary intervention, and highlights the need for effective pharmacological insulin sensitisers to assist in the management of IR and excess BMI in PCOS. Finally, we confirm that SHBG has a strong relationship with IR in PCOS and may be a clinically useful marker of IR and metabolic status in PCOS.

Supplementary data

Supplementary data are available at http://humrep.oxfordjournals.org/.

Authors' roles

S.C., M.L.M., H.J.T. and N.K.S. designed the research, S.C. and M.L.M. performed the literature search, SC and C.S.S. independently reviewed the articles, S.C. performed data extraction. W.G.H. designed the meta-analyses models and W.G.H. and N.K.S. performed the data analysis. S.C., M.L.M., C.S.S., W.G.H., H.J.T. and N.K.S. participated in interpretation. S.C. wrote the manuscript and all authors provided critical review of the manuscript draft and approved the final version.

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Conflict of interest

The authors have no conflict of interests to declare.

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