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ORIGINAL ARTICLE

Effect of metformin by employing 2-hour postload insulin for measuring insulin resistance in Taiwanese women with polycystic ovary syndrome

Huang-tz Ou [a,*], Pei-Chi Chen [a], Meng-Hsing Wu [b,c]

[a] *Institute of Clinical Pharmacy and Pharmaceutical Sciences, College of Medicine, National Cheng Kung University, Tainan, Taiwan*
[b] *Department of Obstetrics and Gynecology, College of Medicine, National Cheng Kung University, Tainan, Taiwan*
[c] *Department of Obstetrics and Gynecology, National Cheng Kung University Hospital, Tainan, Taiwan*

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Background/purpose: Evidence on clinical effectiveness of metformin in ethnic Chinese women with polycystic ovary syndrome (PCOS) remains scarce. Standard diagnostic approaches to identify insulin resistance (IR) cases in PCOS patients might be invasive, labor intensive, and stressful for patients (i.e., euglycemic clamp), or somewhat complicated for clinicians to calculate and monitor in routine practice [i.e., the homeostatic model assessment (HOMA) and quantitative insulin sensitivity check index (QUICKI)]. The aim of this study was to evaluate the clinical effects of metformin in Taiwanese women with PCOS and identify the feasible diagnostic measures of IR for Taiwanese women with PCOS.

Methods: A total of 114 women from a medical center in Taiwan were studied. All were aged between 18 years and 45 years, diagnosed with PCOS according to the Rotterdam criteria, and treated with metformin. Outcome end points were body mass index (BMI) and 2-hour postload glucose and insulin levels from a 75-g oral glucose tolerance test.

Results: BMI in overweight patients were significantly improved with metformin treatment duration ($p < 0.001$). The 2-hour insulin level statistically improved after treatment (before: 80.7 ± 63.9 μIU/mL vs. after: 65.0 ± 60.4 μIU/mL; $p = 0.009$). The improved 2-hour insulin level was significantly greater in IR patients than in non-IR patients. Compared with the 2-hour postload insulin level, the fasting insulin level provided 18.15% sensitivity and 94.12% specificity, the HOMA yielded 40% sensitivity and 70.58% specificity, and the QUICKI achieved 63.63% sensitivity and 11.76% specificity.

Conflicts of interest: The authors have no conflicts of interest relevant to this article.

* Corresponding author. Institute of Clinical Pharmacy and Pharmaceutical Sciences, College of Medicine, National Cheng Kung University, 1, University Road, Tainan 7010, Taiwan.
E-mail address: huangtz@mail.ncku.edu.tw (H.-t. Ou).

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Conclusion: Clinical outcomes in Taiwanese PCOS women were improved with metformin treatment, especially in overweight and IR patients. The 2-hour postload insulin level appears to be a convenient tool for screening IR in Taiwanese patients.

Introduction

Polycystic ovary syndrome (PCOS) is the most common endocrine disorder of reproductive-age women, approximately affecting 8.7—11.9% of such individuals, which depends on diagnostic criteria [i.e., National Institutes of Health (NIH) criteria, Rotterdam consensus criteria, and Androgen Excess Society criteria].[1] According to the Rotterdam criteria, 5.6% of Chinese women suffer from PCOS.[2] The common clinical presentations associated with this syndrome are obesity, menstrual disorders, acne, hirsutism, and alopecia.[3] Around 60—70% of women with PCOS have insulin resistance (IR).[4] Obese women with PCOS have significantly more severe IR levels than nonobese women with PCOS.[5] Because of the high prevalence of IR, women with PCOS are prone to metabolic syndromes,[6] such as obesity, impaired glucose tolerance (IGT), and gestational and Type 2 diabetes, and have an increased risk of cardiovascular diseases.[7]

A biguanide medication, metformin, reduces insulin secretion and hyperinsulinemia, and improves insulin sensitivity, ovary function, and associated metabolic syndromes.[8] Although metformin use for PCOS is still considered "off-label" by the Food and Drug Administration, it is commonly prescribed for women with PCOS,[9] especially for those with IR.[10] Some studies have demonstrated that metformin improves glucose effectiveness,[11] insulin sensitivity, menstrual cyclicity, fertility, and live-birth rates, and reduces clinical hyperandrogenism (i.e., acne),[9,12] whereas another study did not find such clinical benefits of metformin when used for PCOS.[13] However, evidence on clinical effects of metformin in Chinese women with PCOS remains limited. Recently, Li et al's study[14] of 47 metformin-treated PCOS women from China showed significantly lower 2-hour insulin levels in oral glucose tolerance test (OGTT) and an improvement in insulin sensitivity as shown by the significant decrease in the homeostatic model assessment (HOMA) of IR after 6 months of treatment.

Adherence to treatment is a key for ensuring drug response and effectiveness in patients.[15] In previous studies on patients with Type 2 diabetes, patients' adherence with metformin was found to be suboptimal and to decrease with time,[16,17] mostly due to its side effects [i.e., gastrointestinal (GI) intolerance]. Few studies have assessed medication adherence issues in PCOS women.[18,19] Identifying the factors associated with medication adherence is important for developing treatment education and interventions to enhance patients' drug compliance and ensure the effectiveness of treatment in clinical practice.[15] It is thus important for researchers to elucidate the adherence behavior in PCOS women and assess the factors associated with their medication adherence.

Several diagnostic approaches have been applied to assess IR in PCOS women. Some direct measures typically require an intervention (e.g., administrate insulin) to assess the ability of insulin to dispose glucose, including the euglycemic clamp and intravenous glucose tolerance test/OGTT.[20] However, they are usually invasive, labor intensive, and stressful for patients. Surrogate methods assess the fasting insulin level and glucose-to-insulin (G/I) ratio to estimate insulin action.[20] These methods are more rapid, less invasive, and require less time and skill to perform. However, fasting insulin levels and G/I ratios do not accurately reflect insulin sensitivity.[21] The HOMA and the quantitative insulin sensitivity check index (QUICKI) scores can be calculated based on the fasting glucose and fasting insulin concentrations to determine IR. However, they may not be feasible for clinicians to calculate in practice. Saxena et al[22] studied Indian women with PCOS and showed that the 2-hour postglucose-load insulin level from OGTT is a cost-effective, convenient, and reliable marker for IR, especially in resource-constrained developing countries. Future research is thus warranted to determine whether the OGTT is a convenient tool for screening IR in Taiwanese PCOS patients.

The present study therefore aims to (1) examine the clinical effectiveness of metformin in Taiwanese women with PCOS, stratifying the effects by patients' characteristics, namely, body mass index (BMI; overweight vs. normal), glucose tolerance (IGT vs. normal), and insulin sensitivity (IR vs. normal); (2) evaluate patients' adherence to metformin and the factors associated with metformin treatment duration for women with PCOS; and (3) identify a feasible and convenient approach (i.e., fasting insulin, HOMA, QUICKI, 2-hour postload insulin from OGTT) to determine IR in PCOS women in clinical practice.

Materials and methods

A retrospective cohort study was conducted to evaluate the clinical effectiveness of metformin by reviewing medical charts in the National Cheng Kung University Hospital from October 2004 to December 31, 2011. Permission for the study from the Institutional Review Board of the National Cheng Kung University Hospital was obtained before the study commencement (A-ER-103-287). Because this retrospective study analyzed and reported the data anonymously, no informed consent was obtained from the study participants.

Participants

The study cohort was aged between 18 years and 45 years and diagnosed with PCOS during the period from January 1, 2005, to December 31, 2010, according to the Rotterdam

criteria[23] in which PCOS was defined by the presence of at least two of the following three criteria: (1) oligo-anovulation (a cycle length $>$ 35 days or amenorrhoea), (2) clinical hyperandrogenism (hirsutism recorded as modified Ferriman–Gallwey score $\geq$ 6 with/without acne or androgenic alopecia) and/or biochemical hyperandrogenism (total testosterone level $>$ 0.95 ng/mL), and (3) polycystic ovaries ($\geq$ 12 follicles measuring 2–9 mm in diameter, or ovarian volume $>$ 10 mL in at least one ovary). We excluded those (1) diagnosed with similar clinical presentations, including congenital adrenal hyperplasia, hyperprolactinemia, androgen-secreting tumors, Cushing syndrome, abnormal thyroid function, and thyroid disease; (2) diagnosed with diabetes, fasting plasma glucose level of 126 mg/dL or more, 2-hour glucose level of 200 mg/dL or more, or HbA1c greater than 7 before PCOS diagnosis; (3) taking any medications that may influence the insulin or/and glucose level, including metformin and other antidiabetic drugs, or oral contraceptive pills 3 months prior to PCOS diagnosis. All patients were treated with metformin (Glucophage, 500 mg t.i.d. for at least 6 months) after PCOS diagnosis (new users of metformin). Patients who cannot tolerate the immediate-release formulation (i.e., due to GI intolerance side effects) can switch to extended-release (XR) metformin (C.T.L. XR®). Although no study reported similar efficacy between XR and immediate-release metformin in PCOS women, several studies in patients with Type 2 diabetes have shown that XR metformin is as effective as immediate-release metformin to achieve glycemic control in patients newly started on metformin as well as in those switched from the immediate-release formulation.[24–26] Every patient was observed from the initiation of metformin treatment to (1) lost to follow-up, (2) the end of metformin treatment, or (3) the end of 2011. The use of metformin was examined as the length of metformin treatment duration and medication adherence, expressed as the medication possession ratio (MPR). The MPR expresses the percentage of day's supply received divided by a period, implying the percentage of time a patient received a medication (i.e., metformin in this case). The MPR values were calculated by summing day's supply from the first to the last prescription divided by time between the last prescription date plus day's supply and the first prescription date [total days supply/(last fill date − first fill date+ last fill days supply)][27,28] for all patients within a year from the beginning of metformin treatment. The MPR values range from 0 to 1, with higher values indicating better adherence.

Effectiveness outcomes

There were three clinical outcomes of interest, including BMI and 2-hour postload glucose and insulin levels from a standardized 75-g oral OGTT, which were measured every time patients returned for a visit. BMI was estimated as weight divided by height squared. All patients received a 75-g glucose monohydrate in 350 mL water after an 8-hour overnight fasting. A total of 5 mL blood sample was drawn before glucose loading and another 5-mL blood sample was drawn at 120 minutes after the glucose loading. Plasma glucose and insulin concentrations were determined by the glucose oxidase method using a glucose analyzer (Model 2300; YSI, Yellow Springs, OH, USA) and an automated chemiluminescence system (ADVIA Centaur Immunoassay System, Siemens Healthcare Diagnostics, Deerfield, IL, USA), respectively. Other biochemical assessments included complete hormonal evaluation, including serum follicle-stimulating hormone, luteinizing hormone, thyroid-stimulating hormone, prolactin, estradiol (E2), total testosterone, and sex hormone-binding globulin, which were measured on Days 3–5 during patients' menstrual period and analyzed by RIA kit from Diagnostic Products (Los Angeles, CA, USA). The HbA1c level was measured by boronate affinity chromatography (Bio-Rad Laboratories, Inc., Hercules, CA, USA).

Analytic analyses

Descriptive analyses were used to present the demographics of the study population, medication use, and outcomes of interest. Mixed-effect models were applied to assess the effect of metformin (treatment duration and MPR) on the repeated outcomes measure (i.e., BMI) every 3 months in the follow-up. The 2-hour glucose and insulin levels before and after metformin treatment was initiated were evaluated using the paired *t* test. The study population was also stratified by (1) BMI (normal: BMI $<$ 25 kg/m^2 vs. overweight: BMI $\geq$ 25 kg/m^2, and an additional sensitivity analysis: normal: BMI $<$ 27 kg/m^2 vs. overweight: BMI $\geq$ 27 kg/m^2); (2) 2-hour postload glucose level (a cutoff point $\geq$ 140 mg/dL to classify IGT vs. normal); and (3) 2-hour postload insulin level (a cutoff point $\geq$ 35 μIU/mL to classify IR vs. normal) in the OGTT. Although there is no consensus on a cutoff point of the 2-hour insulin level to define IR, Stovall et al[21] showed that the means of the 2-hour postload insulin level for nonoverweight (BMI $<$ 25 kg/m^2) and overweight (BMI $\geq$ 25 kg/m^2) patients were 34.2 μIU/mL and 70.0 μIU/mL, respectively.[21] In addition, Saxena et al[22] used a value of 2-hour insulin level greater than 41 μIU/mL to determine the presence of IR in Indian women with PCOS.[22] We selected a cutoff point of 2-hour insulin level of 35 μIU/mL or more based on the range of 2-hour postload insulin levels in Stovall et al's study[21] (34.2–70.0 μIU/mL) and our laboratory at the National Cheng Kung University Hospital. The low cutoff point value of 2-hour postload insulin (i.e., 35 μIU/mL) we used might reduce the bias of study findings toward a falsely low rate of IR. The difference in difference test was applied to assess the change in glucose (or insulin) levels before and after metformin treatment between subgroups (i.e., IGT and normal patients, IR, and normal patients), with adjustment for the baseline patients' characteristics and medication use [i.e., metformin treatment duration (MPR)]. Furthermore, using the 2-hour insulin level (with a value of $\geq$ 35 μIU/mL defined as IR cases) from the OGTT as a standard to identify IR cases, we estimated the sensitivity and specificity of the fasting insulin (with a threshold $>$ 20 μIU/mL defined as IR[21,29]), HOMA[30] (with a value $\geq$ 2.14 defined as IR[31]), and QUICKI[32] (with a value $\geq$ 0.34 defined as IR[31]). A receiver-operating characteristic (ROC) curve was plotted to compare the HOMA and QUICKI with the OGTT in the prediction of IR cases.

Statistical significance was set to a p value less than 0.05. Statistical Analysis System 9.4 (SAS Institute Inc., Cary, NC, USA) was used for the aforementioned statistical analyses.

Results

A total of 114 patients were included (Figure 1), with an average age of 27.1 years, 37% with BMI of 25 kg/m^2 or more, 38% with IGT, and 66% with IR. All eligible women (n = 114) had baseline examination and had return office visits of at least two times after PCOS diagnosis. Of them, 72 women had complete data on fasting glucose and insulin levels and 2-hour glucose and insulin levels. All study participants started with immediate-release metformin (Glucophage) and then 41% of patients switched to XR metformin (C.T.L. XR®) in follow-up. The detailed baseline characteristics of the study population are presented in Table 1. Supplementary Table S1 online presents the demographics of obese subgroups with various cutoff values of BMI. In multivariate analysis, XR metformin use was significantly associated with treatment duration of metformin; compared with immediate-release metformin users, those with the XR form of metformin treatment had 3.29 months longer metformin treatment (p = 0.003; Table 2). In addition, patients who received the XR form showed better adherence to metformin than those who received the immediate-release form; the average value of MPR in the users of XR form (MPR = 0.89) was significantly higher than that in those of the immediate-release form (MPR = 0.70; $p < 0.001$). Using a cutoff point of MPR of 0.85 or over for stratifying adherence and nonadherence,[33] 87% of patients with XR had MPR values greater than 0.85, whereas only 53% of those with immediate-release form had values of 0.85 or more. Table 3 indicates significant improvement in BMI in overweight and obese women with PCOS, whereas this was not significant in PCOS women with normal weight. Figure 2A and 2B indicate that the 2-hour glucose and insulin levels decreased after metformin treatment. Although the change in the 2-hour glucose level was not statistically significant (baseline: 112.0 ± 27.2 mg/dL vs. 113.0 ± 32.2 mg/dL, p = 0.75; Figure 2A), the improved 2-hour insulin level after metformin treatment was statistically significant (baseline: 80.7 ± 63.9 μIU/mL vs. 65.0 ± 60.4 μIU/mL, p = 0.009; Figure 2B). A plot of 2-hour post-load glucose/insulin ratio was also provided in Supplementary Figure S3. IGT and IR patients had significantly improved 2-hour glucose and insulin levels after metformin

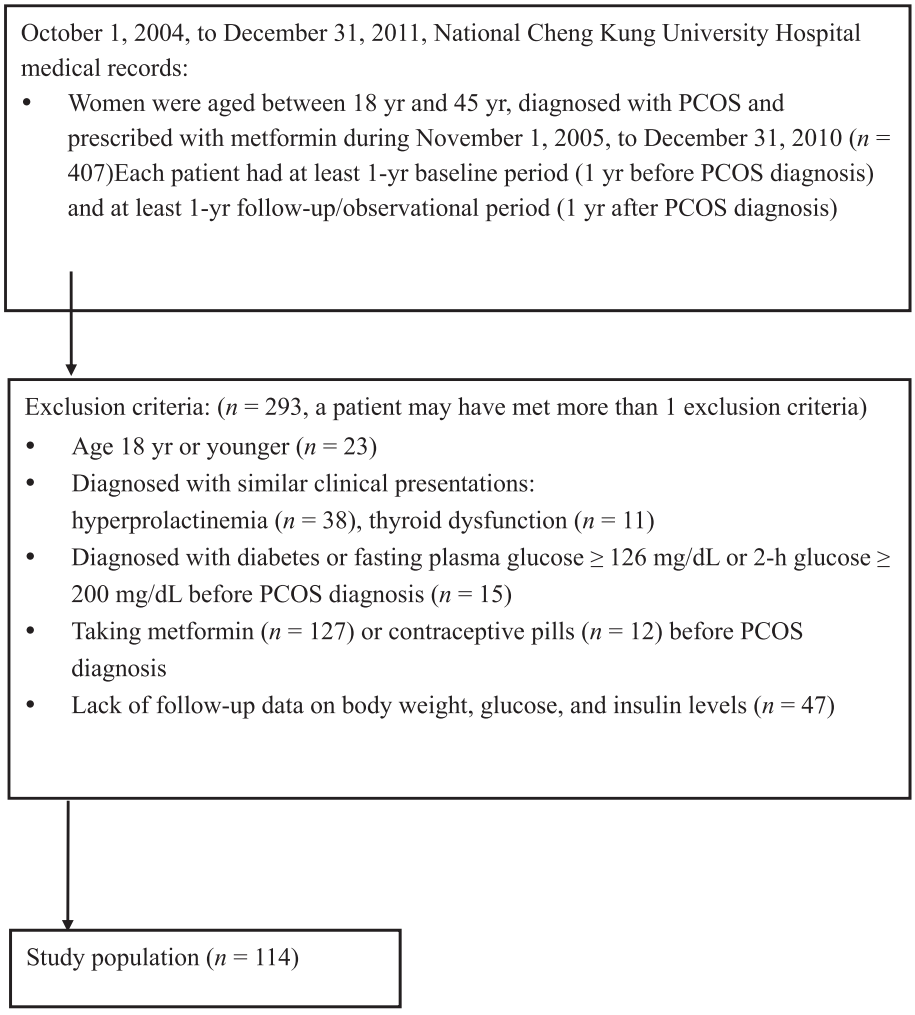

Figure 1 Flowchart of study population selection. PCOS = polycystic ovary syndrome.

Table 1 Demographics of study population.

Characteristics	All ($n = 114$)	Subgroups					
		Nonoverweight (BMI < 25 kg/m^2) ($n = 59$)	Overweight (BMI ≥ 25 kg/m^2) ($n = 55$)	Normal ($n = 71$)	Impaired glucose tolerance ($n = 43$)	Normal ($n = 39$)	Insulin resistance ($n = 75$)
Biometric							
Age (yr)	27.1 ± 4.7	26.7 ± 4.1	27.8 ± 5.2	26.3 ± 4.8	26.9 ± 3.7	25.5 ± 5.1	26.7 ± 4.5
Weight (kg)	64.2 ± 15.5	53.6 ± 6.5	78.6 ± 13.8*	65.9 ± 16.8	70.6 ± 6.2	58.1 ± 11.9	68.7 ± 17.3**
BMI (kg/m^2)	25.1 ± 5.8	21.0 ± 2.2	30.9 ± 4.7*	25.8 ± 6.2	27.5 ± 7.4	23.3 ± 5.4	26.7 ± 0.78**
Systolic BP (mmHg)	118.3 ± 17.7	109.4 ± 10.9	127.5 ± 19.0*	117.7 ± 17.5	130.4 ± 23.9**	117.2 ± 15.9	120.4 ± 19.1
Diastolic BP (mmHg)	72.6 ± 13.0	66.0 ± 8.0	79.9 ± 13.2*	72.0 ± 12.7	82.1 ± 18.8**	68.8 ± 13.5	74.3 ± 13.7
m-FG score	4.1 ± 3.4	3.9 ± 2.8	4.4 ± 4.0	4.9 ± 3.8	1.8 ± 1.3**	6.1 ± 4.8	4.2 ± 3.3
Hormones							
TSH (μIU/mL)	1.98 ± 0.84	1.81 ± 0.70	2.08 ± 0.88	1.97 ± 0.83	2.26 ± 1.00	2.16 ± 0.90	1.98 ± 0.82
LH (mIU/mL)	9.70 ± 5.92	11.17 ± 6.40	7.52 ± 4.03**	8.94 ± 5.21	8.00 ± 5.49	9.66 ± 4.56	8.66 ± 5.36
FSH (mIU/mL)	6.28 ± 2.06	6.14 ± 1.88	6.21 ± 2.12	6.14 ± 1.94	6.08 ± 2.09	6.24 ± 1.72	6.05 ± 2.02
LH/FSH	1.39 ± 0.49	1.67 ± 0.53	1.21 ± 0.27	1.36 ± 0 .43	1.17 ± 0.51	1.48 ± 0.33	1.29 ± 0.46
Estradiol (E2, pg/mL)	52.64 ± 42.48	46.59 ± 31.07	56.81 ± 53.65	50.00 ± 32.27	52.85 ± 22.26	45.99 ± 37.67	51.60 ± 29.07
PRL (ng/mL)	10.31 ± 4.31	10.65 ± 4.33	9.58 ± 4.21	10.50 ± 4.12	8.86 ± 3.67	9.40 ± 2.70	10.60 ± 4.34
TT (ng/mL)	0.54 ± 0.28	0.48 ± 0.25	0.60 ± 0.30**	0.59 ± 0.30	0.62 ± 0.36	0.51 ± 0.26	0.61 ± 0.29
Glycemic parameters							
2-h glucose (mg/dL)	109.6 ± 27.9	102.4 ± 27.5	120.4 ± 27.3**	104.5 ± 17.9	166.1 ± 15.9*	91.4 ± 17.0	117.0 ± 26.7*
2-h insulin (μIU/mL)	72.4 ± 59.5	50.3 ± 32.1	106.1 ± 76.5*	69.2 ± 51.5	159.6 ± 91.3**	18.1 ± 9.5	97.0 ± 61.9*
Clinical presentation							
Biochemical hyperandrogenism	17 (11.4)	4 (6.8)	10 (18.2)	12 (16.9)	2 (20.0)	2 (11.8)	11 (16.9)
Hirsutism	30 (27.0)	13 (28.3)	11 (25.6)	21 (34.4)	0 (0.0)	8 (53.3)	14 (25.9)
Acne	72 (48.3)	29 (49.2)	27 (49.1)	29 (41.4)	5 (50.0)	6 (75.0)	28 (65.1)
Irregular menses	96 (84.2)	45 (76.3)	51 (92.7)	58 (81.7)	38 (88.4)	31 (79.5)	64 (85.3)

Except for clinical presentation [presented as *n* (%)], all other data are presented as mean ± standard deviation. The *n* (%) refers to the number of patients with a given clinical condition (i.e., biochemical hyperandrogenism) and the percentage of patients with a given condition. Patients with irregular menses were treated as oligomenorrhea and amenorrhea cases.

* $p \leq 0.0001$.

** $p \leq 0.05$.

BMI = body mass index; BP = blood pressure; FSH = follicle-stimulating hormone; LH = luteinizing hormone; m-FG = modified Ferriman–Gallwey; PRL = prolactin; TSH = thyroid-stimulating hormone; TT = total testosterone.

Table 2 Factors associated with treatment duration of metformin for women with PCOS.

	Coefficient (standard error)	*p*
Outcome: metformin treatment duration (month)		
Univariate analysis		
Weight (kg)	0.092 (0.032)	0.005
BMI (kg/m^2)	0.259 (0.087)	0.003
Overweight (BMI $\geq$ 25 kg/m^2) (ref. normal weight)	2.264 (1.023)	0.029
Metformin extended-release (ref. immediate-release)	4.068 (1.019)	0.0001
Marital status (ref. married)	2.184 (1.017)	0.033
Pregnancy intention (ref. no pregnancy intention or plan)	2.183 (1.034)	0.036
Multivariate analysis (R^2 = 0.146)		
Metformin extended-release (ref. immediate-release)	3.288 (1.066)	0.003

BMI = body mass index; PCOS = polycystic ovary syndrome; ref. = reference.

treatment than those without IGT and IR, respectively (Table 4). A total of 72 cases had complete values of the fasting glucose, fasting serum insulin (from which the HOMA and QUICKI could be calculated), and 2-hour postload insulin from the OGTT. The ROC curves were 0.562 [95% confidence interval (CI) 0.4842–0.6388], 0.552 (95% CI 0.4236–0.6823), and 0.6230 (95% CI 0.5213–0.7247) for the fasting insulin, HOMA and QUICKI, respectively (see Supplementary Figure S1 online). As compared with the 2-hour postload insulin level, the fasting insulin level provided 18.15% sensitivity and 94.12% specificity, the HOMA had 40% sensitivity and 70.58% specificity, and the QUICKI achieved 63.63% sensitivity and 11.76% specificity.

Discussion

This study showed that metformin significantly improved IR and BMI, especially in overweight and IR women affected by PCOS. Our results support the recommendations advocated by the 2013 Endocrine Society Clinical Guideline,[8] which indicates that for women with PCOS, metformin is recommended for overweight, IR, or IGT cases. In addition, the XR metformin that has lower GI side effects and pill burden (i.e., once-daily dosing), compared with the conventional immediate-release formulation, appeared to enhance patients' adherence to treatment.

Comparison with previous research on women with PCOS

Several international studies recently evaluated the clinical effectiveness of metformin for women with PCOS. Fux Otta et al[34] conducted a randomized, double-blind, and placebo-controlled trial for 4 months to examine the effects of metformin versus a placebo, in addition to diet and exercise, on endocrine and metabolic disturbances in 30 Argentine women with PCOS (20–34 years old), where the definition of PCOS diagnosis followed the NIH criteria. A prospective observational study in India was carried out by Saxena et al[35] on 40 infertile women (aged 18–38 years) with PCOS (confirmed by the Rotterdam criteria). All patients were treated with metformin by 3 months. Pau et al[11] conducted an open-label, interventional study of 36

Table 3 Mixed effect models for assessing the effect of metformin treatment on body mass index.

	All (n = 114)		Subgroup analyses							
			Nonoverweight (BMI < 25 kg/m^2) (n = 59)		Overweight (BMI $\geq$ 25 kg/m^2) (n = 55)		Nonobese (BMI < 27 kg/m^2) (n = 70)		Obese (BMI $\geq$ 27 kg/m^2) (n = 44)	
	Estimate (standard error)	*p*	Estimate (standard error)	*p*	Estimate (standard error)	*p*	Estimate (standard error)	*p*	Estimate (standard error)	*p*
Intercept	25.80 (0.58)	<0.0001	21.06 (0.28)	<0.0001	30.93 (0.64)	<0.0001	21.80 (0.32)	<0.0001	32.22 (0.67)	<0.0001
Metformin treatment duration		0.002		0.63		0.0003		0.28		0.0001
3 mo (ref. baseline)	−0.05 (0.30)	0.88	0.18 (0.35)	0.61	−0.52 (0.50)	0.30	−0.06 (0.31)	0.84	−0.34 (0.58)	0.55
6 mo (ref. baseline)	−0.39 (0.30)	0.19	0.13 (0.35)	0.71	−1.08 (0.50)	0.03	−0.13 (0.30)	0.67	−1.01 (0.59)	0.09
9 mo (ref. baseline)	−0.50 (0.31)	0.11	0.02 (0.35)	0.96	−1.21 (0.51)	0.02	−0.27 (0.30)	0.36	−1.09 (0.62)	0.09
Medication possession ratio	−0.49 (0.32)	0.13	−0.53 (0.38)	0.16	−0.19 (0.54)	0.72	−0.29 (0.33)	0.38	−0.47 (0.63)	0.46

The mixed effect models were adjusted for blood pressures, LH, TT, 2-hour glucose and 2-hour insulin levels, which were statistically different between nonoverweight and overweight patients at baseline (shown in Table 1).
BMI = body mass index; LH = luteinizing hormone; TT = total testosterone.

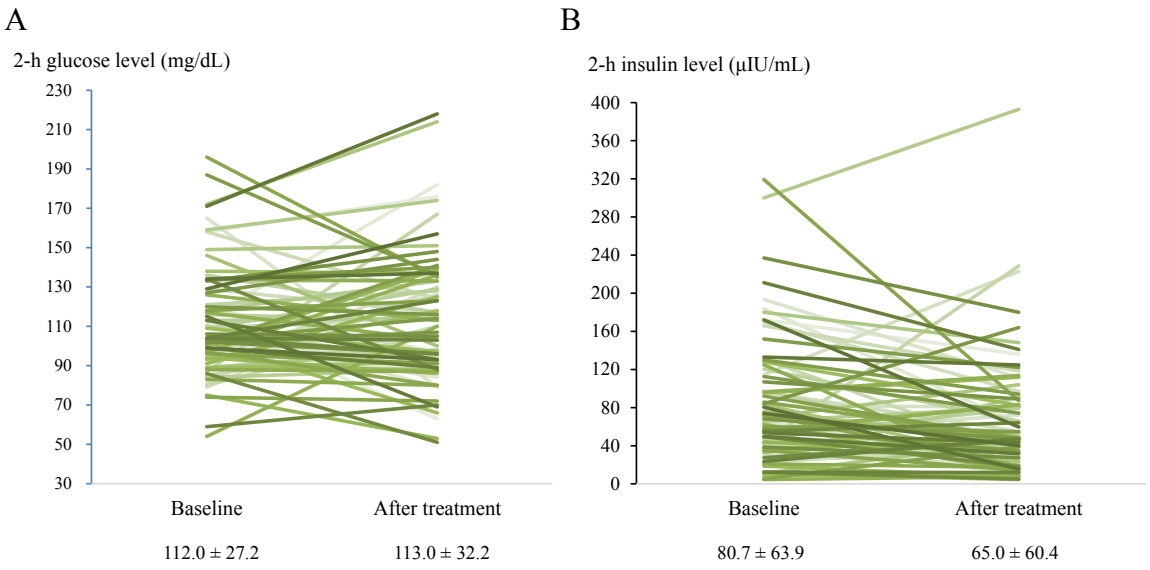

Figure 2 Change in 2-hour postload (A) glucose and (B) insulin levels before and after 6 months of metformin treatment. (A) The plot shows 53.1% of polycystic ovary syndrome (PCOS) patients with decreased glucose levels, 45.7% with increased glucose levels, and 1.2% with no change. The difference in the mean value of the 2-hour glucose level before and after metformin treatment was not statistically significant ($p = 0.75$). In addition, our subgroup analysis indicated that 22 of the 43 impaired glucose tolerance patients (50%) became normal, whereas seven of the 71 patients (9.9%) without impaired glucose tolerance at the baseline showed impaired glucose tolerance after treatment (see Supplementary Figure S2A online). (B) The plot shows 68.3% of PCOS patients with decreased insulin levels and 31.7% with increased insulin levels. The difference in the mean value of the 2-hour insulin level before and after metformin treatment was statistically significant ($p = 0.009$). Our subgroup analysis further indicates that 21 of the 75 insulin resistance patients (27.7%) became normal, whereas 16 of the 39 patients (41.2%) without insulin resistance at the baseline showed insulin resistance after treatment (see Supplementary Figure S2B online).

Table 4 Effect of metformin on 2-hour postload glucose and insulin levels within and between groups.

2-h glucose level (mg/dL)				
Subgroups	Baseline (mean ± SD)	After treatment (mean ± SD)	DID within group (mean ± SD, *p*)	DID between two groups (mean ± SD, *p*)
Normal ($n = 71$)	104.3 ± 18.3	107.7 ± 26.3	3.7 ± 3.4, $p = 0.29$	−21.4 ± 9.9, $p = 0.03$
Impaired glucose tolerance ($n = 43$)	166.1 ± 15.9	150.7 ± 45.5	−17.7 ± 9.3, $p = 0.06$	
2-h insulin level (μIU/mL)				
Subgroups	Baseline (mean ± SD)	After treatment (mean ± SD)	DID within group (mean ± SD, *p*)	DID between two groups (mean ± SD, *p*)
Normal ($n = 39$)	18.3 ± 9.6	28.3 ± 23.4	11.8 ± 13.0, $p = 0.37$	−34.7 ± 14.7, $p = 0.02$
Insulin resistance ($n = 75$)	97.1 ± 61.9	74.6 ± 63.4	−22.9 ± 6.6, $p = 0.0008$	

The DID analyses were adjusted for baseline blood pressure, metformin treatment duration, and adherence.
DID = difference-in-difference; SD = standard deviation.

American women with PCOS (diagnosed using the NIH criteria) from an academic medical center in the United States and assessed the effects of metformin before and after 12 weeks of metformin treatment. Li et al[14] reported the clinical effects of 6-month metformin treatment on 47 PCOS women from China.

In terms of body weight, the current study and those mentioned earlier[11,14,35] all showed that the patients' BMI significantly decreased after metformin treatment, but Fux Otta et al[34] did not find a similar trend. Regarding the 2-hour postload glucose level from OGTT, both Fux Otta et al[34] and Pau et al[11] reported a significant glycemic change after metformin treatment; however, our study and that of Li et al[14] found a nonsignificant improvement, whereas Saxena et al[35] did not find a similar trend. Our subgroup analysis further showed that over 50% of IGT patients became normal after metformin treatment, whereas 9.9% of PCOS women without IGT turned into IGT (see Supplementary Figure S2A online). These results indicate that metformin might be more effective in IGT cases than in those without IGT.

Moreover, our study results and those of Saxena et al[35] and Li et al[14] demonstrated that the 2-hour insulin level significantly lowered after metformin treatment. Our

subgroup analysis further indicated that 28% of IR patients became normal after treatment, whereas 41% of PCOS women without IR turned into IR (see Supplementary Figure S2B online). However, the study results of Pau et al[11] and Fux Otta et al[34] showed that metformin treatment was not associated with improved insulin sensitivity for PCOS women. The difference in metformin effect on insulin sensitivity for PCOS women across studies might be explained by different diagnostic techniques, cutoff points for defining IR, and study population. Using the intravenous glucose tolerance test, results in women with PCOS have demonstrated no change in insulin sensitivity after 12 weeks to 3 months of metformin treatment.[11,36,37] By contrast, both Saxena et al's[35] results based on a cutoff point of 2-hour insulin level over 41 μIU/mL and our study results based on a cutoff value of 2-hour insulin level of 35 μIU/mL or more to determine IR showed improved insulin sensitivity after metformin treatment. In Li et al's study,[14] improved insulin sensitivity was found, as indicated by the significant decrease in the HOMA score after 6 months of metformin treatment. In addition, using a euglycemic hyperinsulinemic clamp was found to improve insulin sensitivity.[38,39] Moreover, Li et al's study[14] targeted Chinese PCOS women from China and our study focused on Taiwanese PCOS women, whereas the other aforementioned studies[11,35,36–39] were from other countries/ethnicity. Therefore, we suggest that future study should be carried out to investigate any variations in metformin efficacy for PCOS women due to different measuring approaches and countries/ethnicity.

Metformin adherence among women with PCOS

Most previous research examined metformin adherence in diabetes patients,[16,17] but only little information is available about metformin use behavior in PCOS women. Li et al's study[19] of 99 infertile PCOS women showed that 23% of patients had good drug compliance as measured by the Morisky–Green test, a patient self-reported medication adherence questionnaire. Their study also showed that PCOS women who had adverse drug reactions or experienced inconvenient medical treatment (i.e., pill burden) were likely to exhibit noncompliance. The present study showed that, as measured by MPR, a pill count-based medication adherence measure, PCOS women had satisfactory medication adherence (mean of MPR > 0.85) as compared with previously reported metformin adherence in chronic disease patients (e.g., diabetes[16,17]). This is in part because women with PCOS tend to be young. Most of our participants had a high educational level and might have had good health literacy in terms of their self-care and/or health-seeking behaviors. Moreover, we found that XR metformin prolonged patients' treatment duration. It has been reported that the side effects of metformin (e.g., GI intolerance) may limit its use in diabetic patients.[16,17] As compared with conventional immediate-release metformin, the XR metformin formulation reduces GI side effects and allows once-daily dosing, reducing the pill burden for patients and enhancing patients' medication adherence, thus ensuring the efficacy of treatment.[24–26,40] Consistent with these findings, Li et al's study[19] showed that PCOS patients who had adverse drug reactions or experienced inconvenient medical treatment (i.e., pill burden) were likely to exhibit noncompliance. In addition, less adverse drug events might lead to better drug compliance in PCOS women.[18] Therefore, XR metformin is an option for PCOS women who experience GI intolerance due to the immediate-release formulation or for those with polypharmacy issues; additionally, this may improve their adherence to metformin.

Approaches for diagnosing IR in Taiwanese women with PCOS

Early detection of IR cases is important because such patients are prone to the risks of metabolic syndrome and Type 2 diabetes.[6] Because of lack of uniform diagnostic measure to assess IR, the incidence of IR in PCOS women is difficult to estimate,[22] leading to discrepancies in IR rates that varied with diagnostic approaches,[41] inadequate therapy, and misleading interpretations from the results of treatment. Available diagnostic methods are expensive and might not be convenient in practice. For routine practice, a feasible method to determine IR in PCOS women is needed.[41] We compared the 2-hour postload insulin level from OGTT with commonly used approaches, including fasting insulin, HOMA, and QUICKI in diagnosing IR in Taiwanese PCOS women. We sought to determine whether the 2-hour insulin level from OGTT is a convenient tool for screening IR cases in PCOS women. Our results imply that the 2-hour insulin level from OGTT might be a feasible and convenient method for screening IR in Taiwanese PCOS women. Our additional analysis showed that 75.4%, 69.4%, 37.5%, and 14.9% of patients in this study ($n = 72$) were classified as having IR based on the 2-hour postload insulin level from the OGTT, QUICKI, HOMA, and fasting insulin results, respectively (see Supplementary Table S2 online). As compared with gold standard (i.e., the euglycemic clamp), the OGTT is less invasive and less expensive, requires less time and skill, thus reducing costs of repeat visits and is easier to apply in clinical practice. Therefore, our study supports the results of Saxena et al's[22] study that the 2-hour postload insulin level from the OGTT appears to be a feasible maker to diagnose IR in PCOS women, especially in resource-restricted health care settings.

Potential limitations

Several limitations of this study need to be acknowledged. First, this was an observational study without a comparison group (i.e., without metformin treatment, oral contraceptives), and therefore, the results only note the improvement after metformin treatment. Second, the present study only used objective medication adherence measures based on patients' "refill pattern" (i.e., the length/duration of metformin treatment, MPR) to determine patients' adherence. However, it is still uncertain whether patients truly took the medication or even used it appropriately. Subjective medication use/adherence measures such as the Morisky medication adherence questionnaire should be applied in future studies to control for potential impact of patients' medication behavior on clinical outcomes of

interest. Third, all participants were from one medical center in Taiwan. Future research is warranted to verify our findings using a larger sample from different regions with various education and socioeconomic statuses of Chinese population. Fourth, Saxena et al[22] used a cutoff value of 2-hour postload insulin level greater than 41 μIU/mL to determine IR in Indian PCOS women, whereas our study selected a value of 2-hour insulin level of 35 μIU/mL or more to determine IR in Taiwanese PCOS women. However, until now no study has examined what cutoff point of 2-hour postload insulin should be used to diagnose IR in PCOS women. Therefore, if a single 2-hour postglucose insulin level appears to be a reliable indicator of IR in PCOS women, it is critical for further research to identify a specific and sensitive cutoff point of 2-hour postload insulin for determining IR. In addition, further research is needed to evaluate if 2-hour postload insulin level is a dedicated marker for monitoring treatment responses and determine whether there is a liner relationship between 2-hour postload insulin level and metformin treatment.

Conclusion

This study of Taiwanese women with PCOS showed that metformin significantly improved BMI and insulin sensitivity, especially for overweight, IGT, and IR cases. Metformin could thus be considered as a suitable treatment for Taiwanese PCOS patients who are overweight, IGT, or IR. The 2-hour postload insulin level from OGTT appears to be a promising approach for screening IR in ethnic Taiwanese women with PCOS. Thus, therapeutic interventions (i.e., metformin) can be initiated, reducing future risk of developing diabetes.

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Appendix A. Supplementary data

Supplementary data related to this article can be found at http://dx.doi.org/10.1016/j.jfma.2016.02.001.

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