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Sustainability of 8% weight loss, reduction of insulin resistance, and amelioration of atherogenic-metabolic risk factors over 4 years by metformin-diet in women with polycystic ovary syndrome

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Abstract

In 74 women with polycystic ovary syndrome, treated for 4 years with metformin (MET) and diet, we prospectively assessed whether, and to what degree, weight loss, reduction of insulin resistance, and amelioration of coronary heart disease risk factors could be sustained. We hypothesized that response to MET-diet would not differ by pretreatment body mass index (BMI) classes <25 (normal), ≥25 to <30 (overweight), ≥30 to <40 (obese), and ≥40 (extremely obese).

		Median		Percent body weight loss category			
	BMI	Weight (kg)	% Weight loss	5%-10%	10%-15%	15%-20%	≥20%
Entry	33.8	90.5					
1-y treatment	30.1	81	8.1	32%	21%	14%	4%
2-y treatment	30.2	82	8.5	27%	23%	11%	8%
3-y treatment	30.8	83	7.8	23%	19%	12%	10%
4-y treatment	30.0	83	8.2	16%	18%	15%	8%

Metformin-diet was successful in producing stable ~8% weight reduction for all 4 years (trend $P < .0001$). Percentage of reductions in weight on MET-diet was significant ($P < .05$) and did not differ among the 3 highest BMI categories (≥40, ≥30 to <40, ≥25 to <30), but were not significant in the normal-weight category (BMI, <25). On MET-diet, median homeostasis model assessment of insulin resistance (HOMA-IR) was 33% lower than entry at 1 year, 50% at 2 years, 51% at 3 years, and 50% at 4 years (trend, $P < .0001$). On MET-diet, median low-density lipoprotein cholesterol (LDL-C) was 6% lower than entry at year 1, 6% at year 2, 7% at year 3, and 11% at year 4 (trend $P < .0001$). On MET-diet, median high-density lipoprotein cholesterol (HDL-C) was 3% higher than entry at year 2, 8% higher at year 3, and 11% higher at year 4 (trend $P < .0001$). Percentage of reductions in HOMA-IR, LDL-C, triglyceride, and systolic blood pressure, and increments in HDL-C did not differ ($P > .1$) in the 4 BMI categories. By stepwise regression, weight loss was a significant ($P \leq .01$) positive explanatory variable for reduction in HOMA-IR for all 4 follow-up years. Metformin-diet in women with polycystic ovary syndrome effectively and safely reduces weight and LDL-C while raising HDL-C, and maintains these outcomes stable over 4 years.

1. Introduction

Polycystic ovary syndrome (PCOS) affects approximately 6% of adult whites [1], 13% of Hispanics [2], and 8% of African Americans [3]. The PCOS phenotype commonly includes rapid weight gain from menarche to young adulthood [4], biochemical and/or clinical hyperandrogenism (severe acne, hirsutism), oligoamenorrhea, polycystic ovaries, and usually, but not always, insulin resistance (IR) [3-14]. During the passage from adolescence to young adulthood, phenotypic and clinical abnormalities of PCOS worsen, and are commonly accompanied by infertility,

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frequent first-trimester miscarriage, and increased likelihood of gestational diabetes [4,10,11,15,16].

In adult women with PCOS, there is an increase in subclinical atherosclerosis as diagnosed by increased carotid intima-media thickness (IMT) [5,6]. An actual increase in ischemic myocardial infarction and stroke has not been demonstrated by prospective studies of well-characterized populations of women with PCOS [7]. However, adult women with PCOS appear to be at increased risk for atherosclerotic cardiovascular disease, attributed in part to their hyperandrogenemia and to their metabolic syndrome that includes central obesity, hyperglycemia, type 2 diabetes mellitus, hyperlipidemia, hypofibrinolysis, hypertension, and hyperinsulinemia [8-10].

Low-energy [11,12], high-protein, low-carbohydrate diet and intensive exercise [13,14], as well as metformin (MET)-diet, benefit adults with PCOS [15-19]. Lifestyle modification alone, MET alone, and their combination improve weight, endocrinopathy, and menstrual abnormalities in PCOS [14,18,20-22]. Moran et al [23] gave a 6-kJ diet, either low protein (55% carbohydrate, 15% protein) or high protein (45% carbohydrate, 30% protein), to women with PCOS and after 16 weeks reported 7% weight loss in the low-protein diet and 8.2% weight loss in the high-protein diet. Moran et al [24] have reported that appetite regulation, as measured by subjective short-term hunger and satiety and ghrelin homeostasis, may be impaired in PCOS. Although short-term energy restriction successfully improves endocrine function and leads to weight loss in PCOS [23], longer-term energy restriction is often unsuccessful [25].

Metformin augments diet-induced weight loss in women with PCOS [26]. Pasquali et al [26] gave a 5.02- to 5.858-kJ diet (50% carbohydrate, 20% protein) to women with PCOS and then added MET 1700 mg/d, reporting a 8.7% weight loss on MET-diet vs 4.9% on placebo-diet after 28 weeks on therapy. Comparing 12 months MET-diet in women with PCOS in the top and bottom quintiles for homeostasis model assessment of IR (HOMA-IR) [27], we reported that weight fell by 7% in both quintile groups ($P < .0001$). Insulin, IR, and insulin secretion fell in the top quintile insulin-resistant group by 60%, 64%, and 39% (all $P < .0001$), with smaller reductions in the bottom quintile group by 18%, 13% ($P > .05$ for both), and 22% ($P < .01$), respectively [27]. Metformin-diet metabolic effects were much more marked in women in the top vs the bottom quintile for IR [27].

Rapid weight gain from menarche, which characterizes PCOS [4,28], has major cardiovascular health consequences in adulthood as well as adverse behavioral effects during adolescence [29]. Childhood low-density lipoprotein cholesterol (LDL-C) and BMI predict carotid IMT in young adults [30]. Increased carotid IMT is observed among overweight (BMI, ≥95th percentile) children who became obese (BMI, ≥30 kg/m^2) adults [31]. However, IMT was not increased among overweight children who were not obese in adulthood or among thinner children who became obese adults [31]. Cardiovascular risk in young adulthood is closely related to the degree of adiposity as early as 13 years of age [32].

In 74 women with PCOS, treated for 4 years with MET and diet, we prospectively assessed whether, and to what degree, weight loss, reduction of IR, and amelioration of coronary heart disease risk factors could be sustained. We hypothesized that response to MET-diet would not differ by pretreatment BMI classes <25 (normal), ≥25 to <30 (overweight), ≥30 to <40 (obese), and ≥40 (extremely obese) [33].

2. Materials and methods

2.1. Study design, cases

The study was carried out following a protocol approved by the Jewish Hospital (Cincinnati, OH) institutional review board, with signed informed consent.

The diagnosis of PCOS was made based on the revised 2003 Rotterdam European Society for Human Reproduction and Embryology/American Society for Reproductive Medicine (ESHRE/ASRM) consensus criteria [34], with cases meeting 2 or more of the following 3 criteria after exclusion of other pathologies (pituitary insufficiency, persistent hyperprolactinemia, congenital adrenal hyperplasia, etc):

1. oligoamenorrhea or anovulation;
2. clinical [35] and/or biochemical signs of hyperandrogenism;
3. polycystic ovaries.

Additional exclusion criteria in our study were premenarchal status at study entry, serum creatinine of more than 1.5 mg/dL, type 1 diabetes mellitus, type 2 diabetes mellitus on pharmacologic therapy, hypothyroidism, pregnancy, and concurrent use of estrogen-progestin oral contraceptives.

Cases were referred for evaluation by their pediatricians, gynecologists, or by other family members with PCOS. All postmenarchal women who fulfilled the Rotterdam PCOS definition [34] were included in the study without known selection bias. At the initial outpatient visit, weight and height were measured, and blood was obtained after an overnight fast for measurement of thyroxine, thyrotropin, hepatic and renal function, complete blood count, serum insulin, glucose, testosterone, estradiol, progesterone, lipid profile, and plasminogen activator inhibitor activity [28,36,37]. Homeostasis model assessment of IR was calculated as per Matthews et al [38]. After a 5-minute resting period, seated blood pressure [39] was obtained by a single observer.

Subjects were initially instructed one on one by registered dietitians, with follow-up reinstruction once per year. Dietary energy (26% protein, 44% carbohydrate [42% of carbohydrate complex], 30% fat) was targeted to 6.28 kJ to 7.53 kJ/d if BMI was less than 25 (normal), or to 5.02 kJ to 6.28 kJ/d if BMI was 25 or more (overweight) [33]. Metformin was given along with diet (2550 mg/d).

Table 1
Body weight, HOMA-IR, triglycerides, HDL-C, LDL-C, SBP, and DBP (median and median of percent changes) in 74 women with PCOS at pretreatment and on follow-up at years 1 to 4 on MET-diet

	Pretreatment	On MET-diet (year 1)	On MET-diet (year 2)	On MET-diet (year 3)	On MET-diet (year 4)	Significance of changes*
Weight (kg)	90.5	81	82	83	83	Pretreatment higher than on MET-diet, $P < .0001$
		−8%	−9%	−8%	−8%	Stable during years 1-4, $P = .63$
IR	3.48	1.99	1.97	1.94	1.71	Pretreatment higher than on MET-diet, $P < .0001$
		−33%	−50%	−51%	−50%	Stable during years 1-4, $P = .16$
TG (mg/dL)	123	90	93	92	101	Pretreatment higher than on MET-diet, $P < .0001$
		−17%	−20%	−16%	−11%	Stable during years 1-4, $P = .72$
HDL-C (mg/dL)	46	45	47	50	50	Pretreatment did not differ from on MET-diet year 1, $P = .65$
		−3%	+3%	+8%	+11%	Year 1 was lower than years 2-4, $P = .0004$ Stable during years 2-4, $P = .18$
LDL-C (mg/dL)	119	107	106	105	103	Pretreatment higher than on MET-diet, $P < .0001$
		−6%	−6%	−7%	−11%	Stable during years 1-4, $P = .85$
SBP (mm Hg)	124	120	121	122	118	Pretreatment higher than on MET-diet, $P < .0001$
		−7%	−5%	−5%	−7%	Stable during years 1-4, $P = .47$
DBP (mm Hg)	82	74	75	76	76	Pretreatment higher than on MET-diet, $P < .0001$
		−7%	−6%	−3%	−5%	Year 1 was lower than years 3 and 4, $P < .036$ Stable during years 2-4, $P = .37$

TG indicates triglycerides.
* Repeated-measures ANOVA.

Outpatient visits were repeated after an overnight fast every 2 months for 4 years, with measurement of weight, insulin, glucose, testosterone, estradiol, progesterone, lipid profile, and systolic blood pressure (SBP) and diastolic blood pressure (DBP). At yearly follow-up, renal and hepatic function was remeasured. At each visit, medication use was recorded. No quantitative measures of dietary adherence were obtained. Our goal was to assess sustainability of weight loss, reduction of HOMA-IR, and amelioration of atherogenic-metabolic risk factors over 4 years of therapy with MET-diet, and to determine if response to MET-diet differed by BMI class.

2.2. Statistical methods

All statistical evaluations were done using SAS (SAS/STAT software, 9.1, SAS Institute, Cary NC). Measurements taken at pretreatment baseline were compared with measurements taken at years 1 to 4 on MET-diet by the mixed-model repeated-measures analysis of variance adjusted for baseline values for each parameter. This allowed analysis for trend over time, and between measures at baseline, and years 1, 2, 3, and 4 (Table 1). Body mass index category was added as a class variable to the mixed model (Fig. 1) to assess whether response to MET-diet differed by BMI category (≥40 [extremely obese], ≥30 but <40 [obese], ≥25 but <30 [overweight], <25 [normal] [33]). To assess whether response to MET-diet differed by 4-year completion of the study protocol vs noncompletion, we added completion category as a class variable to the mixed model.

Stepwise regression models were run for changes at years 1, 2, 3, and 4 with the dependent variables being changes in atherosclerosis risk-metabolic syndrome variables including weight, IR, triglyceride (TG), LDL-C, high-density lipoprotein cholesterol (HDL-C), SBP, and DBP. Explanatory variables included age, race, response variables at pretreatment, pretreatment weight, MET dose, duration on MET, as well as change in weight and/or change in IR (Table 1). In the stepwise regression models with change in triglyceride or HDL-C as dependent variable, change in HDL-C or triglycerides was added, respectively, to the explanatory variable list.

With $\alpha = .05$ and 80% power, assuming, from our previous experience, weight loss on MET-diet of 7% ± 11%

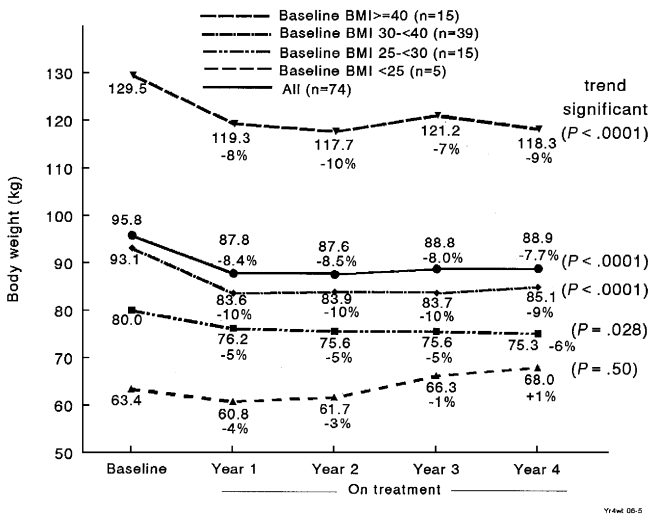

Fig. 1. Mean pretreatment entry body weight (kg), mean body weight at years 1, 2, 3, and 4 on MET-diet, and mean percent change in weight during therapy in the full cohort (solid line). Mean pretreatment entry body weight (kg) and mean body weight at years 1, 2, 3, and 4 on MET-diet compared with pretreatment entry after categorization [33] by pretreatment BMI as normal (<25), overweight (≥25-30), obese (≥30-40), and severely obese (≥40).

(SD), and compared with an assumed 2% or less weight loss without MET-diet, the sample size should be 32 or more.

3. Results

3.1. Cases

The 74 women in the current report came from a group of 1002 women with PCOS referred to our center for diagnosis and treatment of PCOS who met the Rotterdam diagnostic criteria [34]. Of the 1002 women, 314 were initially studied less than 4 years ago and did not have long enough follow-up to be eligible for the current 4 years or more follow-up study, leaving 688 eligible women. Of these 688, 414 had only an initial evaluation without follow-up MET-diet therapy, leaving 274 eligible women. Of these 274 women, 89 had follow-up for ≥1 but <2 years, 42 for ≥2 but <3 years, and 21 for ≥3 but <4 years, leaving 122 women with 4 years or more of follow-up. Of these 122 women, 74 with complete measures at pretreatment baseline and years 1, 2, 3, and 4 comprised the current cohort, whereas 48 did not have complete data over 4 consecutive years. Comparisons were made against the 200 women (follow-up: ≥1 but <2 years [n = 89], ≥2 but <3 [n = 42], ≥3 but >4 [n = 21], ≥4 [n = 48] with incomplete data for 4 years) to assess for selection bias in the 74 studied women. These comparisons included pretreatment age, BMI, glucose, HOMA-IR, race, and percentage of weight loss, and reduction of HOMA IR on MET-diet.

The 74 women with complete 4-year data did not differ ($P > .1$) from the 200 women with incomplete data for any pretreatment characteristics or for change on MET-diet in HOMA-IR and weight.

By selection, the 74 women with PCOS met the Rotterdam diagnostic criteria [34] and had complete 4-year follow-up data. There were 70 white women, 1 African American, and 3 other. Mean ± SD and median age were 31 ± 9 and 32 years; mean ± SD and median BMI, 34.9 ± 7.6 and 33.8 kg/m^2; and mean ± SD and median weight, 95.8 ± 21.6 and 90.5 kg. At study entry, 5 (7%) of women had BMI of less than 25 (normal), 15 (20%) were overweight (BMI, 25-30), 39 (53%) were obese (BMI, ≥30-40), and 15 (20%) were severely obese (BMI, ≥40) [33].

Mean ± SD and median fasting HOMA-IR were 4.50 ± 3.65 and 3.48; mean ± SD and median triglyceride, 140 ± 85 and 123 mg/dL; mean ± SD and median LDL-C, 118 ± 30 and 119 mg/dL; and mean ± SD and median HDL-C, 47 ± 11 and 46 mg/dL.

Mean ± SD and median MET dose were 2395 ± 476 and 2550 mg/d. Adherence to MET was excellent, in part because only branded Glucophage (Bristol-Myers Squibb, Princeton, NJ) was used because it has a much less marked adverse gastrointestinal side effect profile than the generic.

Serial, 4-year determinations of fasting glucose, renal, and hepatic function revealed no adverse effects of diet-MET.

Despite pretreatment severe obesity, MET-diet was successful in producing stable ~8% weight reduction for all 4 years (decreasing trend $P < .0001$) (Table 1, Fig. 1). In women with baseline BMI of ≥40 (extreme obesity), ≥30 to <40 (obese), and ≥25 to <30 (overweight) on MET-diet, there were significant decreasing trends in weight (Fig. 1). In the small group of women with BMI of less than 25 (normal weight), there was no significant trend over time in weight (Fig. 1). Percentage of reductions in weight was significant in the groups with BMI of ≥40, ≥30 to <40, and ≥25 to <30 and did not differ ($P > .1$) in the 3 highest BMI categories. After the first year's reduction in weight on MET-diet, for the full cohort of 74 women, there were no differences ($P \geq .5$) in weight between years 1 to 4 of follow-up (Table 1).

On MET-diet at year 1, median HOMA-IR (1.99) was 33% lower than entry levels (3.48), 50% lower at 2 years, 51% lower at 3 years, and 50% lower at 4 years (decreasing trend, $P < .0001$) (Table 1). After the first year's reduction in HOMA-IR on MET-diet, for the full cohort of 74 women, there were no differences ($P > .15$) in HOMA-IR between years 1 to 4 of follow-up (Table 1). Percentage of reductions in HOMA-IR did not differ ($P > .1$) in the 4 BMI categories.

On MET-diet at year 1, median LDL-C (107 mg/dL) was 6% lower than entry levels (119 mg/dL), 6% lower at year 2, 7% lower at year 3, and 11% lower at year 4 (decreasing trend, $P < .0001$) (Table 1). After the first year's reduction in LDL-C on MET-diet, for the full cohort of 74 women, there were no differences ($P = .9$) in LDL-C between years 1 to 4 of follow-up (Table 1). Percentage of reductions in LDL-C did not differ ($P > .1$) in the 4 BMI categories.

On MET-diet at year 1, median HDL-C (45 mg/dL) was slightly, but not significantly, lower than entry levels (46 mg/dL, $P = .65$), and then rose during year 2 to 47 mg/dL, 3% higher than levels at baseline ($P = .077$). High-density lipoprotein cholesterol was 8% higher than entry at year 3 and 11% at year 4 (increasing trend, $P < .0001$) (Table 1). Percentage of increments in HDL-C did not differ ($P = .4$) in the 4 BMI categories.

On MET-diet at year 1, median triglyceride (90 mg/dL) was 17% lower than entry levels (123 mg/dL), 20% lower at year 2, 16% lower at year 3, and 11% lower at year 4 (decreasing trend, $P < .0001$) (Table 1). After the first year's reduction in triglyceride on MET-diet, for the full cohort of 74 women, there were no differences ($P = .7$) in triglyceride between years 1 to 4 of follow-up (Table 1). Percentage of reductions in triglycerides did not differ ($P = .9$) in the 4 BMI categories.

On MET-diet at year 1, median SBP (120 mm Hg) was 7% lower than entry levels (124 mm Hg), 5% lower at year 2, 5% lower at year 3, and 7% lower at year 4 (decreasing trend, $P < .0001$) (Table 1). After the first year's reduction in SBP on MET-diet, for the full cohort of 74 women, there were no differences ($P = .47$) in SBP between years 1 to

4 of follow-up (Table 1). Percentage of reductions in SBP did not differ ($P = .69$) in the 4 BMI categories.

On MET-diet at year 1, median DBP (74 mm Hg) was 7% lower than entry levels (82 mm Hg), 6% lower at year 2, 3% lower at year 3, and 5% lower at year 4 (decreasing trend, $P < .0001$) (Table 1). After the first year's reduction in DBP on MET-diet, for the full cohort of 74 women, there were no differences ($P = .4$) in DBP between years 2 to 4 of follow-up (Table 1). Percentage of reductions in DBP was larger in the BMI category of ≥30 to <40 ($P = .019$) than in the other 3 categories, which did not differ.

By stepwise regression, with change in weight as the dependent variable, and pretreatment weight, age, race, change in IR, and MET dose and duration as explanatory variables, for all 4-year follow-ups, the greater the reduction in IR on MET-diet, the greater the reduction in weight (partial R^2 = 7%-25%, $P < .05$), (Table 2).

In a stepwise regression model with change in IR as the dependent variable and MET dose, treatment duration, pretreatment weight and IR, age, race, and change in weight on therapy as explanatory variables, for all 4-year follow-ups, the higher the entry IR, the greater the decrement in IR on MET-diet (partial R^2 for entry IR ranged from 61% to 81%, $P < .0001$) (Table 2). The greater the decrement in weight on treatment, the greater the reduction in IR (partial R^2 ranging from 2% to 9%, $P < .04$) (Table 2).

Decrements in triglycerides were related throughout the 4-year follow-up period to entry triglycerides (partial R^2 ranging from 9% to 34%, $P < .006$) (Table 2). Decrements in triglycerides were also related to decrements in weight (partial R^2 ranging from 8% to 14%, $P < .007$) (Table 2).

Decrements in LDL-C were related to entry LDL-C for all 4 years, with partial R^2 ranging from 31% to 50% ($P < .0001$), and to decrements in weight for year 1 (Table 2).

Increments in HDL-C were inversely related to entry HDL-C for all 4 years, with partial R^2 ranging from 9% to 32% ($P \leq .01$), and positively related to decrements in weight for years 1, 2, and 4 (partial R^2 ranging from 6% to 8%) (Table 2). The higher the entry weight, the smaller the increments in HDL-C (partial R^2 ranging from 7% to 11%) (Table 2).

Decrements in SBP were related to entry SBP, with partial R^2 ranging from 35% to 56%, $P < .0001$ (Table 2). At years 1 and 3, the greater the decrement in weight, the greater the decrement in SBP. The greater the entry weight, the smaller was the reduction in SBP on MET-diet (partial R^2 ranging from 7% to 24%) (Table 2).

Decrements in DBP were related to entry DBP with R^2 ranging from 29% to 52% ($P < .0001$) (Table 1). For all 4 years, the greater the decrement in weight, the greater the decrement in DBP (Table 2). The higher the entry weight, the smaller was the decrement in DBP on MET-diet (partial R^2 ranging from 13% to 24%) (Table 2).

Table 2
Significant explanatory variables for changes in 74 women with PCOS during 4 years on MET-diet from pretreatment entry in weight, IR, triglycerides, HDL-C, LDL-C, SBP, and DBP

Dependent Variable	1 y		2 y		3 y		4 y	
	Explanatory variable	Partial R^2, P	Explanatory variable	Partial R^2, P	Explanatory variable	Partial R^2, P	Explanatory variable	Partial R^2, P
Decrease in WT	↓ in IR+	8%, .021	↓ in IR+	13%, .0023	↓ in IR+	7%, .029	↓ in IR+	25%, <.0001
Decrease in IR	Entry IR+	61%, <.0001	Entry IR+	70%, <.0001	Entry IR+	81%, <.0001	Entry IR+	61%, <.0001
	↓ in WT+	2%, .040	↓ in WT+	5%, .0007	↓ in WT+	4%, <.0001	↓ in WT+	9%, <.0001
	Entry WT−	2%, .013	Entry WT−	1%, .047	Entry WT−	4%, <.0001	Entry WT−	4%, .0015
	Age +	2%, .034			Age+	1%, .019		
Decrease in TG	Entry TG+	19%, .0002	↓ in WT+	11%, .0059	Entry TG+	34%, <.0001	Entry TG+	18%, .0004
	Duration−	7%, .013	Entry TG+	9%, .0090	↓ in WT+	10%, .0009	↓ in WT+	14%, .0005
	↓ in WT+	8%, .0074					Entry WT−	5%, .028
	Entry WT−	4%, .037					↓ in IR+	7%, .0089
Increase in HDL-C	Entry HDL-C−	9%, .011	Entry HDL-C−	18%, .0002	Entry HDL-C−	30%, <.0001	Entry HDL-C−	32%, <.0001
	Entry WT−	7%, .020	↓ in WT+	8%, .0084	Entry WT−	7%, .0075	↓ in WT+	7%, .0088
	↓ in WT+	6%, .025	Entry WT−	11%, .0011	Duration+	5%, .023	Entry WT−	8%, .0035
Decrease in LDL-C	Entry LDL-C+	31%, <.0001	Entry LDL-C+	41%, <.0001	Entry LDL-C+	50%, <.0001	Entry LDL-C+	47%, <.0001
	↓ in WT+	13%, .0002						
Decrease in SBP	Entry SBP+	56%, <.0001	Entry SBP+	36%, <.0001	Entry SBP+	35%, <.0001	Entry SBP+	38%, <.0001
	Entry WT−	11%, .0001	Entry WT−	7%, .021	Entry WT−	24%, <.0001	Entry WT−	19%, <.0001
	MET dose+	3%, .019			↓ in WT+	6%, .0060		
	↓ in WT+	3%, .025			Age−	6%, .0036		
					Duration +	3%, .026		
Decrease in DBP	Entry DBP+	52%, <.0001	Entry DBP+	29%, <.0001	Entry DBP+	34%, <.0001	Entry DBP+	29%, <.0001
	Entry WT−	14%, <.0001	Entry WT−	17%, .0002	Entry WT−	24%, <.0001	Entry WT−	13%, .0016
	↓ in WT+	3%, .032	↓ in WT+	8%, .0049	↓ in WT+	9%, .0005	↓ in WT+	5%, .047
	MET dose+	3%, .018			Age−	4%, .011		
					Duration+	4%, .0067		

WT indicates weight; ↓ in, decrease in; ↑ in, increase in; +, positive association with dependent variable; −, inverse association with dependent variable.

4. Discussion

If IR and compensatory hyperinsulinemia are central to the pathophysiology of PCOS [14,23,26,28,40-52], then the success of long-term MET-diet therapy in PCOS should be closely related to reduction in IR and reduction in hyperinsulinemia. In the current study, on MET-diet, there were significant reductions in HOMA-IR at each year of follow-up, ranging from 33% after 1 year to 50% at year 4. Reduction in IR was a significant independent variable associated with reduction in weight at all 4-year follow-ups. Moreover, a major independent determinant of change in IR on MET-diet was entry-pretreatment IR, accounting for 61% of the variance in the decrement in IR at year 1, 70% at year 2, 81% at year 3, and 61% at year 4. Hence, the greater pretreatment IR, the greater was the reduction in IR on MET-diet, such that MET-diet would optimally benefit women with higher pretreatment IR. In a similar fashion, MET-diet therapy would provide more benefit for women with higher pretreatment LDL-C, triglyceride, SBP, and DBP, and lower HDL-C.

In the current study, MET-diet was equally effective in the 3 heaviest BMI groups in regard to weight loss, and equally effective in all 4 BMI groups for changes in IR, LDL-C, HDL-C, TG, and SBP. These findings are congruent with those of Harborne et al [53] who gave MET 1.5 or 2.55 g to women with PCOS who were obese (BMI, 30-37) and morbidly obese (BMI, $\geq$37), reporting significant weight loss in both groups and a dose response for weight loss in the obese group only. Our findings were in contrast to those of Ehrmann et al [54] in obese women with PCOS (mean BMI, 39), where 2.55 g/d MET failed to improve hyperinsulinemia and androgen excess.

Because, in the current study, only 5 (7%) of 74 women had normal BMI (<25), our assessments of the effects of MET-diet in normal-weight women are necessarily limited by small numbers. Although women with pretreatment BMI of less than 25 differed from the other 3 BMI categories for weight loss (did not have significant weight loss on MET-diet), they did not differ from the other 3 (heavier) BMI categories for percent change on MET-diet in IR, LDL-C, HDL-C, triglycerides, or SBP. Women with BMI of less than 25 did not differ from those with BMI of $\geq$25 to <30 or $\geq$40 for change in DBP on MET-diet, but had smaller decrements in DBP on MET-diet than women with BMI of $\geq$30 to <40. Thus, excepting weight loss, women with BMI of less than 25 shared the other benefits of MET-diet equally with those women who were overweight, obese, or extremely obese. Nestler and Jakubowicz [48] have reported that MET treatment reduces hyperandrogenemia in lean women. We [27] have previously reported that women with PCOS in the bottom pretreatment quintile for IR (the leanest at pretreatment) nevertheless experienced significant metabolic and menstrual benefits. In nonobese women with PCOS, Sahin et al [55] reported significant decrements in weight, free testosterone, and measures of IR. In nonobese (mean BMI, 21.4) adolescents with anovulatory hyperandrogenism given 1.275 g MET, Ibanez et al [56] reported that 78% ovulated within 6 months. Maciel et al [57] reported that nonobese women with PCOS had better response to MET 1.5 g for 6 months than obese women, with decrements in insulin, testosterone, and androstenedione.

Relatively short-term studies in women with PCOS have shown that hypoenergetic diet alone for 16 weeks [23] will produce 7% to 8.2% weight loss and hypoenergetic diet plus 1700 mg MET for 28 weeks will produce 8.7% weight loss compared with 4.9% on diet-placebo [26]. However, longer-term energy restriction is often unsuccessful in women with PCOS [25]. Our study is encouraging in as much as it reveals that weight loss of ~8% was maintained over 4 years by MET-diet, along with reduction in IR.

Although our study was not placebo-controlled, and is small, reductions in body weight can be compared with those in the much larger, placebo-controlled study of obese patients (PCOS not specified) receiving orlistat (360 mg/d) for 4 years [58]. Before treatment, orlistat study participants [58] had BMI of 30 or more. Of orlistat-treated patients, only 52% completed treatment compared with 34% of placebo recipients ($P < .0001$). Mean weight loss after 4 years was significantly greater with orlistat (5.8 vs 3.0 kg with placebo, $P < .001$). By comparison, in the current study, mean weight loss on MET-diet at 4 years was 7.5 kg, and the median percentage of weight loss was 8.2%.

Beyond weight loss and reduction in IR, long-term treatment with MET-diet may be antiatherogenic [28,41,59-61] by virtue of reductions of LDL-C and increments in HDL-C, as in the current study. To the extent that PCOS is associated with higher risk of atherosclerosis [62-64], reduction of weight, IR, LDL-C, triglyceride, SBP, and DBP, and increments in HDL-C should protect against atherosclerosis [65] and should have promise in primary prevention of type 2 diabetes mellitus [66-71].

A major limitation of the current study is that it does not allow an independent assessment of diet effect alone [23,25]; this could be observed in a MET-diet vs placebo-diet comparison [26]. This restricts our conclusions beyond that MET-diet results in a sustained ~8% weight loss over 4 years, accompanied (at year 4) by reductions in HOMA-IR (50%), LDL-C (11%), TG (11%), SBP (7%), DBP (5%), and increments in HDL-C (11%). Independent assessment of MET effect is not possible, given the present study design. Although the 74 women with complete 4-year data did not differ for pretreatment characteristics or follow-up changes in HOMA-IR or weight loss on MET-diet with the 200 women with incomplete 4-year follow-up, it is possible that our cohort of 74 women may represent a self-selected group of MET-diet achievers. However, in women with PCOS, long-term diet alone is not very successful, either for weight loss or improvement of endocrine function [25,26,72], whereas, in the current study, MET-diet offered 4-year success. Better agents for weight loss are needed in

PCOS. Although median pretreatment weight (90.5 kg) was successfully reduced after 4 years on MET-diet to 83 kg (median reduction 8.2%, $P < .0001$), the women remained obese (median BMI, 30.0).

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References

[1] Knochenhauer ES, Key TJ, Kahsar-Miller M, et al. Prevalence of the polycystic ovary syndrome in unselected black and white women of the southeastern United States: a prospective study. J Clin Endocrinol Metab 1998;83:3078-82.

[2] Goodarzi MO, Quinones MJ, Azziz R, et al. Polycystic ovary syndrome in Mexican-Americans: prevalence and association with the severity of insulin resistance. Fertil Steril 2005;84:766-9.

[3] Azziz R, Woods KS, Reyna R, et al. The prevalence and features of the polycystic ovary syndrome in an unselected population. J Clin Endocrinol Metab 2004;89:2745-9.

[4] Glueck CJ, Dharashivkar S, Wang P, et al. Obesity and extreme obesity, manifest by ages 20-24 years, continuing through 32-41 years in women, should alert physicians to the diagnostic likelihood of polycystic ovary syndrome as a reversible underlying endocrinopathy. Eur J Obstet Gynecol Reprod Biol 2005;122:206-12.

[5] Vryonidou A, Papatheodorou A, Tavridou A, et al. Association of hyperandrogenemic and metabolic phenotype with carotid intima-media thickness in young women with polycystic ovary syndrome. J Clin Endocrinol Metab 2005;90:2740-6.

[6] Lakhani K, Hardiman P, Seifalian AM. Intima-media thickness of elastic and muscular arteries of young women with polycystic ovaries. Atherosclerosis 2004;175:353-9.

[7] Legro RS. Polycystic ovary syndrome and cardiovascular disease: a premature association? Endocr Rev 2003;24:302-12.

[8] Macut D, Micic D, Cvijovic G, et al. Cardiovascular risk in adolescent and young adult obese females with polycystic ovary syndrome (PCOS). J Pediatr Endocrinol Metab 2001;14(Suppl 5):1353-9 [discussion 1365].

[9] Glueck CJ, Papanna R, Wang P, et al. Incidence and treatment of metabolic syndrome in newly referred women with confirmed polycystic ovarian syndrome. Metabolism 2003;52:908-15.

[10] Lo JC, Feigenbaum SL, Yang J, et al. Epidemiology and adverse cardiovascular risk profile of diagnosed polycystic ovary syndrome. J Clin Endocrinol Metab 2006;91:1357-63.

[11] Crave JC, Fimbel S, Lejeune H, et al. Effects of diet and metformin administration on sex hormone–binding globulin, androgens, and insulin in hirsute and obese women. J Clin Endocrinol Metab 1995;80:2057-62.

[12] Moran LJ, Noakes M, Clifton PM, et al. Short term energy restriction (using meal replacements) improves reproductive parameters in polycystic ovary syndrome. Asia Pac J Clin Nutr 2004;13:S88.

[13] Pasquali R, Antenucci D, Casimirri F, et al. Clinical and hormonal characteristics of obese amenorrheic hyperandrogenic women before and after weight loss. J Clin Endocrinol Metab 1989;68:173-9.

[14] Hoeger KM, Kochman L, Wixom N, et al. A randomized, 48-week, placebo-controlled trial of intensive lifestyle modification and/or metformin therapy in overweight women with polycystic ovary syndrome: a pilot study. Fertil Steril 2004;82:421-9.

[15] Velazquez EM, Mendoza S, Hamer T, et al. Metformin therapy in polycystic ovary syndrome reduces hyperinsulinemia, insulin resistance, hyperandrogenemia, and systolic blood pressure, while facilitating normal menses and pregnancy. Metabolism 1994;43:647-54.

[16] Velazquez E, Acosta A, Mendoza SG. Menstrual cyclicity after metformin therapy in polycystic ovary syndrome. Obstet Gynecol 1997;90:392-5.

[17] Velazquez EM, Mendoza SG, Wang P, et al. Metformin therapy is associated with a decrease in plasma plasminogen activator inhibitor–1, lipoprotein(a), and immunoreactive insulin levels in patients with the polycystic ovary syndrome. Metabolism 1997; 46:454-7.

[18] Fleming R, Hopkinson ZE, Wallace AM, et al. Ovarian function and metabolic factors in women with oligomenorrhea treated with metformin in a randomized double blind placebo-controlled trial. J Clin Endocrinol Metab 2002;87:569-74.

[19] Morin-Papunen LC, Koivunen RM, Ruokonen A, et al. Metformin therapy improves the menstrual pattern with minimal endocrine and metabolic effects in women with polycystic ovary syndrome. Fertil Steril 1998;69:691-6.

[20] Stamets K, Taylor DS, Kunselman A, et al. A randomized trial of the effects of two types of short-term hypocaloric diets on weight loss in women with polycystic ovary syndrome. Fertil Steril 2004; 81:630-7.

[21] Crosignani PG, Colombo M, Vegetti W, et al. Overweight and obese anovulatory patients with polycystic ovaries: parallel improvements in anthropometric indices, ovarian physiology and fertility rate induced by diet. Hum Reprod 2003;18:1928-32.

[22] Tolino A, Gambardella V, Caccavale C, et al. Evaluation of ovarian functionality after a dietary treatment in obese women with polycystic ovary syndrome. Eur J Obstet Gynecol Reprod Biol 2005; 119:87-93.

[23] Moran LJ, Noakes M, Clifton PM, et al. Dietary composition in restoring reproductive and metabolic physiology in overweight women with polycystic ovary syndrome. J Clin Endocrinol Metab 2003;88:812-9.

[24] Moran LJ, Noakes M, Clifton PM, et al. Ghrelin and measures of satiety are altered in polycystic ovary syndrome but not differentially affected by diet composition. J Clin Endocrinol Metab 2004;89:3337-44.

[25] Norman RJ, Davies MJ, Lord J, et al. The role of lifestyle modification in polycystic ovary syndrome. Trends Endocrinol Metab 2002;13:251-7.

[26] Pasquali R, Gambineri A, Biscotti D, et al. Effect of long-term treatment with metformin added to hypocaloric diet on body composition, fat distribution, and androgen and insulin levels in abdominally obese women with and without the polycystic ovary syndrome. J Clin Endocrinol Metab 2000;85:2767-74.

[27] Goldenberg N, Glueck CJ, Loftspring M, et al. Metformin-diet benefits in women with polycystic ovary syndrome in the bottom and top quintiles for insulin resistance. Metabolism 2005;54:113-21.

[28] Glueck CJ, Wang P, Fontaine R, et al. Metformin to restore normal menses in oligo-amenorrheic teenage girls with polycystic ovary syndrome (PCOS). J Adolesc Health 2001;29:160-9.

[29] Trent M, Austin SB, Rich M, et al. Overweight status of adolescent girls with polycystic ovary syndrome: body mass index as mediator of quality of life. Ambul Pediatr 2005;5:107-11.

[30] Li S, Chen W, Srinivasan SR, et al. Childhood cardiovascular risk factors and carotid vascular changes in adulthood: the Bogalusa Heart Study. JAMA 2003;290:2271-6.

[31] Freedman DS, Dietz WH, Tang R, et al. The relation of obesity throughout life to carotid intima-media thickness in adulthood: the Bogalusa Heart Study. Int J Obes Relat Metab Disord 2004;28: 159-66.

[32] Steinberger J, Moran A, Hong CP, et al. Adiposity in childhood predicts obesity and insulin resistance in young adulthood. J Pediatr 2001;138:469-73.

[33] Flegal KM, Carroll MD, Ogden CL, et al. Prevalence and trends in obesity among US adults, 1999-2000. JAMA 2002;288:1723-37.

[34] The Rotterdam ESHRE/ASRM-Sponsored PCOs Consensus Work Group. Revised 2003 consensus on diagnostic criteria and long-term

health risks related to polycystic ovary syndrome. Fertil Steril 2004; 81:19-25.
[35] Ferriman D, Gallwey JD. Clinical assessment of body hair growth in women. J Clin Endocrinol Metab 1961;21:1440-7.
[36] Glueck CJ, Streicher P, Wang P. Treatment of polycystic ovary syndrome with insulin-lowering agents. Expert Opin Pharmacother 2002;3:1177-89.
[37] Glueck CJ, Wang P, Goldenberg N, et al. Pregnancy outcomes among women with polycystic ovary syndrome treated with metformin. Hum Reprod 2002;17:2858-64.
[38] Matthews DR, Hosker JP, Rudenski AS, et al. Homeostasis model assessment: insulin resistance and beta-cell function from fasting plasma glucose and insulin concentrations in man. Diabetologia 1985;28:412-9.
[39] Ford ES, Giles WH, Dietz WH. Prevalence of the metabolic syndrome among US adults: findings from the third National Health and Nutrition Examination Survey. JAMA 2002;287:356-9.
[40] Glueck CJ, Wang P, Fontaine R, et al. Metformin-induced resumption of normal menses in 39 of 43 (91%) previously amenorrheic women with the polycystic ovary syndrome. Metabolism 1999;48:511-9.
[41] Harborne L, Fleming R, Lyall H, et al. Metformin or antiandrogen in the treatment of hirsutism in polycystic ovary syndrome. J Clin Endocrinol Metab 2003;88:4116-23.
[42] Ibanez L, de Zegher F. Flutamide-metformin plus ethinylestradiol-drospirenone for lipolysis and antiatherogenesis in young women with ovarian hyperandrogenism: the key role of metformin at the start and after more than one year of therapy. J Clin Endocrinol Metab 2005;90:39-43.
[43] Moghetti P, Castello R, Negri C, et al. Metformin effects on clinical features, endocrine and metabolic profiles, and insulin sensitivity in polycystic ovary syndrome: a randomized, double-blind, placebo-controlled 6-month trial, followed by open, long-term clinical evaluation. J Clin Endocrinol Metab 2000;85:139-46.
[44] Sepilian V, Nagamani M. Adiponectin levels in women with polycystic ovary syndrome and severe insulin resistance. J Soc Gynecol Investig 2005;12:129-34.
[45] Spranger J, Mohlig M, Wegewitz U, et al. Adiponectin is independently associated with insulin sensitivity in women with polycystic ovary syndrome. Clin Endocrinol (Oxf) 2004;61:738-46.
[46] Nestler JE, Powers LP, Matt DW, et al. A direct effect of hyperinsulinemia on serum sex hormone–binding globulin levels in obese women with the polycystic ovary syndrome. J Clin Endocrinol Metab 1991;72:83-9.
[47] Nestler JE, Jakubowicz DJ, de Vargas AF, et al. Insulin stimulates testosterone biosynthesis by human thecal cells from women with polycystic ovary syndrome by activating its own receptor and using inositolglycan mediators as the signal transduction system. J Clin Endocrinol Metab 1998;83:2001-5.
[48] Nestler JE, Jakubowicz DJ. Lean women with polycystic ovary syndrome respond to insulin reduction with decreases in ovarian P450c17 alpha activity and serum androgens. J Clin Endocrinol Metab 1997;82:4075-9.
[49] Nestler JE. Insulin resistance syndrome and polycystic ovary syndrome. Endocr Pract 2003;9(Suppl 2):86-9.
[50] Nestler JE. Role of hyperinsulinemia in the pathogenesis of the polycystic ovary syndrome, and its clinical implications. Semin Reprod Endocrinol 1997;15:111-22.
[51] Baillargeon JP, Jakubowicz DJ, Iuorno MJ, et al. Effects of metformin and rosiglitazone, alone and in combination, in nonobese women with polycystic ovary syndrome and normal indices of insulin sensitivity. Fertil Steril 2004;82:893-902.
[52] Baillargeon JP, Iuorno MJ, Nestler JE. Insulin sensitizers for polycystic ovary syndrome. Clin Obstet Gynecol 2003;46: 325-40.
[53] Harborne LR, Sattar N, Norman JE, et al. Metformin and weight loss in obese women with polycystic ovary syndrome: comparison of doses. J Clin Endocrinol Metab 2005;90:4593-8.
[54] Ehrmann DA, Cavaghan MK, Imperial J, et al. Effects of metformin on insulin secretion, insulin action, and ovarian steroidogenesis in women with polycystic ovary syndrome. J Clin Endocrinol Metab 1997;82:524-30.
[55] Sahin I, Serter R, Karakurt F, et al. Metformin versus flutamide in the treatment of metabolic consequences of non-obese young women with polycystic ovary syndrome: a randomized prospective study. Gynecol Endocrinol 2004;19:115-24.
[56] Ibanez L, Valls C, Ferrer A, et al. Sensitization to insulin induces ovulation in nonobese adolescents with anovulatory hyperandrogenism. J Clin Endocrinol Metab 2001;86:3595-8.
[57] Maciel GA, Soares J, Junior M, Alves da Motta EL, et al. Nonobese women with polycystic ovary syndrome respond better than obese women to treatment with metformin. Fertil Steril 2004;81:355-60.
[58] Torgerson JS, Hauptman J, Boldrin MN, et al. XENical in the prevention of diabetes in obese subjects (XENDOS) study: a randomized study of orlistat as an adjunct to lifestyle changes for the prevention of type 2 diabetes in obese patients. Diabetes Care 2004;27:155-61.
[59] Jayagopal V, Kilpatrick ES, Holding S, et al. Orlistat is as beneficial as metformin in the treatment of polycystic ovarian syndrome. J Clin Endocrinol Metab 2005;90:729-33.
[60] De Leo V, la Marca A, Petraglia F. Insulin-lowering agents in the management of polycystic ovary syndrome. Endocr Rev 2003;24: 633-67.
[61] Chou KH, von Eye H, Corleta E, et al. Clinical, metabolic and endocrine parameters in response to metformin in obese women with polycystic ovary syndrome: a randomized, double-blind and placebo-controlled trial. Horm Metab Res 2003;35:86-91.
[62] Talbott EO, Zborowskii JV, Boudraux MY. Do women with polycystic ovary syndrome have an increased risk of cardiovascular disease? Review of the evidence. Minerva Ginecol 2004; 56:27-39.
[63] Talbott EO, Zborowski JV, Rager JR, et al. Evidence for an association between metabolic cardiovascular syndrome and coronary and aortic calcification among women with polycystic ovary syndrome. J Clin Endocrinol Metab 2004;89:5454-61.
[64] Talbott EO, Guzick DS, Sutton-Tyrrell K, et al. Evidence for association between polycystic ovary syndrome and premature carotid atherosclerosis in middle-aged women. Arterioscler Thromb Vasc Biol 2000;20:2414-21.
[65] Tamsma JT, Jazet IM, Beishuizen ED, et al. The metabolic syndrome: a vascular perspective. Eur J Intern Med 2005;16:314-20.
[66] Jermendy G. Can type 2 diabetes mellitus be considered preventable? Diabetes Res Clin Pract 2005;68(Suppl 1):S73-81.
[67] Ramachandran A, Snehalatha C, Mary S, et al. The Indian Diabetes Prevention Programme shows that lifestyle modification and metformin prevent type 2 diabetes in Asian Indian subjects with impaired glucose tolerance (IDPP-1). Diabetologia 2006;1-9.
[68] Laakso M. Prevention of type 2 diabetes. Curr Mol Med 2005; 5:365-74.
[69] Padwal R, Majumdar SR, Johnson JA, et al. A systematic review of drug therapy to delay or prevent type 2 diabetes. Diabetes Care 2005;28:736-44.
[70] Anderson Jr DC. Pharmacologic prevention or delay of type 2 diabetes mellitus. Ann Pharmacother 2005;39:102-9.
[71] Hess AM, Sullivan DL. Metformin for prevention of type 2 diabetes. Ann Pharmacother 2004;38:1283-5.
[72] Kiddy DS, Hamilton-Fairley D, Bush A, et al. Improvement in endocrine and ovarian function during dietary treatment of obese women with polycystic ovary syndrome. Clin Endocrinol (Oxf) 1992; 36:105-11.