Research Article

Age, Body Mass Index, and Waist-to-Hip Ratio Related Changes in Insulin Secretion and Insulin Sensitivity in Women with Polycystic Ovary Syndrome: Minimal Model Analyses

Mirjana Šumarac-Dumanović, Danica Stamenković-Pejković, Danka Jeremić, Janko Dumanović, Vesna Mandić-Marković, Miloš Žarković, and Dragan Mičić

1School of Medicine, University of Belgrade, Belgrade, Serbia
2Clinic for Endocrinology, Diabetes and Diseases of Metabolism, Clinical Center of Serbia, Belgrade, Serbia
3Obstetrics and Gynaecology Clinic Narodni Front, Belgrade, Serbia
4Department of Medical Sciences, Serbian Academy of Sciences and Arts, Belgrade, Serbia

Correspondence should be addressed to Mirjana Šumarac-Dumanović; msumaracdumanovic@gmail.com

Received 24 October 2020; Revised 21 February 2022; Accepted 7 April 2022; Published 18 May 2022

Academic Editor: Arturo Bevilacqua

Copyright © 2022 Mirjana Šumarac-Dumanović et al. This is an open access article distributed under the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

Insulin resistance is believed to be an integral component of the polycystic ovary syndrome (PCOS). Beta (β) cell dysfunction is also found in PCOS. In the study, we determined the influence of age, body mass index (BMI), and waist-to-hip ratio (WHR) on insulin response to oral glucose load (OGTT) and on insulin sensitivity (Si) and β-cell function in young women with PCOS. One hundred fourteen patients with PCOS and 41 controls with normal basal plasma glucose were studied. A 75-g OGTT was performed to determine glucose tolerance and insulin response. Insulin sensitivity and β-cell function were studied using a modified frequently sampled IV glucose tolerance test (FISGTT) to determine the acute insulin response (AIRG), as well as Si by minimal model analysis. Si was decreased in PCOS women (2.49 ± 0.18 vs. 3.41 ± 0.36, p < 0.05), but no difference in AIRG existed between the PCOS and control group (75.1 ± 4.6 vs. 63.4 ± 4.6, p < 0.05). BMI and WHR correlated negatively with Si (r = −0.43; r = −0.289, p < 0.001, respectively), but not with AIRG (r = 0.116; r = −0.02, p > 0.05, respectively). Increasing age correlated negatively with AIRG (r = −0.285, p < 0.001). There was a significant interaction between disease (PCOS), BMI, and WHR on Si as well as between age and PCOS on AIRG. Patients below the age of 25 with PCOS showed enhanced AIRG (89.5 ± 7.1 vs. 65.1 ± 6.7, p < 0.05) and decreased Si (2.43 ± 0.25 vs. 4.52 ± 0.62, p < 0.05) compared to age-matched controls. In conclusion, these data suggest that not all patients with PCOS have basal and stimulated hyperinsulinemia, insulin resistance, and impaired glucose tolerance. Based on these data in young PCOS subjects, the development of insulin resistance and T2DM may be prevented with appropriate treatment strategies.

1. Introduction

Polycystic ovary syndrome (PCOS) is one of the most frequent endocrinopathies in women of reproductive age [1]. The diagnosis of PCOS is suggested by the findings of hyperandrogenism and infertility [2]. Insulin resistance is a coexisting characteristic of this disorder in many mature patients with established PCOS, but its role in the pathogenesis of PCOS is unclear [3–5]. While insulin resistance may be a factor in the development of PCOS, the associated failure of pancreatic β-cell function may also be an important determinant of impaired glucose tolerance or type 2 diabetes (T2DM). Knockout experiments confirm that type 2 diabetes is a “2-hit” disease, in which insulin resistance is necessarily accompanied by a β-defect, preventing the compensatory up-regulation of insulin secretion [6]. The prevalence of impaired glucose tolerance (IGT) and T2DM is increased in PCOS [6]. The clinical characteristics of PCOS, including insulin resistance, have been studied in adolescent persons with this disorder. It is postulated that
the disorder begins at menarche, and some characteristics change with age [7]. A previous study [8] suggested that adolescents with PCOS are severely insulin resistant, compared with a control group matched for body composition and abdominal obesity. Middle-aged PCOS women have been observed to have an increased prevalence of T2DM when compared to an age-matched control population [9].

Thus, the aim of the present study was to investigate the oral glucose tolerance test (OGTT), insulin sensitivity (Si), and acute insulin response (AIRG) during a frequently sampled glucose tolerance test (FSIGTT) as well as the interactions of age, body mass index (BMI), and waist-to-hip ratio (WHR) with the PCOS disorder in a relatively young group of women with PCOS.

2. Materials and Methods

2.1. Subjects. 114 women with PCOS and 41 years of age and BMI-matched healthy women were referred consecutively to the outpatient clinic of the Clinic for Endocrinology, Diabetes, and Diseases of Metabolism for clinical hyperandrogenism and/or menstrual dysfunction. PCOS was diagnosed according to the Rotterdam workshop criteria, i.e., the presence of at least two among the three following features: clinical and/or biochemical hyperandrogenism, chronic oligoanovulation, and polycystic ovary morphology (PCOM), after exclusion of secondary causes [2]. Appropriate tests were used to confirm the absence of specific diseases of the adrenal, ovary or pituitary, such as nonclassic 21-hydroxylase deficiency, hyperprolactinemia, or androgen-secreting neoplasms [1]. No women were taking medications which could potentially interfere with the evaluations carried out in the study. Moreover, patients had not received oral contraceptives, insulin-sensitizing agents, antiandrogens, or glucocorticoids in the six months prior to the investigation. BMI was calculated as body weight/height (kg/m²), and WHR was determined by measuring the waist and hip circumferences at the largest circumference. A BMI of 25 kg/m² was used to define the boundary between overweight and nonoverweight subjects. All the investigated subjects had normal fasting plasma glucose (≤ 5.6 mmol/l) except one PCOS patient who had fasting plasma glucose of 5.8 mmol/L. All controls had normal glucose tolerance based on 2-hr plasma glucose levels during OGTT [10]. Women with PCOS were studied in their follicular phase of the menstrual cycle or were amenorhoeic for more than three months, while control women were tested during their follicular phase. The local human investigation committee approved the study protocol, and all participants gave informed consent.

2.2. Protocol. Oral glucose tolerance test (OGTT) and intravenous glucose tolerance test (IVGTT) with frequent blood sampling (FSIGTT) were conducted on separate occasions in all subjects. Tests were performed after 3 days on a 300 g per day carbohydrate diet and after an overweight fast of 10 hr. Blood samples for plasma glucose and plasma insulin were drawn at baseline and every 30 min for 2 hr, after a 75-g glucose load. The modified IVGTT (FISGTT) was also performed after overnight fasting, according to the previously published procedures [11, 12]. After an overnight fast, catheters were placed in a forearm vein and a hand vein of the contralateral arm. Basal samples were collected for glucose and insulin at −15, −10, −5, and −1 min. Glucose (300 mg/kg) was injected as a bolus at time 0 over 1 min and flushed with saline to ensure complete delivery. After 20 min, 0.05 IU/kg of short-acting insulin (Actrapid HM, NovoNordisk) was injected. Blood samples were drawn at 2, 3, 4, 5, 6, 8, 10, 12, 14, 16, 19, 22, 23, 24, 25, 27, 30, 40, 50, 60, 70, 90, 100, 120, 140, 160, and 180 min for measuring plasma glucose and insulin levels. Glucose was measured in a Beckman glucose analyser, using the glucose oxidase method. Insulin (mU/L) and testosterone (nmol/L) (basal) were measured by radioimmunoassay (RIA INEP, Zemun).

2.3. Data Analysis. The area under the glucose (AUCG) and insulin (AUCI) response curves during OGTT was calculated by the standard trapezoidal rule. The insulin sensitivity index (Si) was calculated by a minimal model analysis using the MINMOD computer program [13]. An acute insulin response (AIRG) was calculated as the mean increase in insulin levels calculated from 2 to 10 min of IVGTT. The disposition index was calculated according to the following formula: $DI = SI \times AIR$. The glucose tolerance to IV glucose load (Kg) was calculated according to the standard procedures [12]. Data were compared using $T$-test, and age and BMI multivariate probability distribution was compared using the peacock test. Multivariate probability distributions were not different between the groups ($p > 0.305$).

Best subset regression was done using the leaps package in order to estimate the best predictor of BMI as well as WHR [14]. Data are presented as the mean ± SEM. Comparisons between groups were performed using the general factorial analysis of the covariance model, controlling for the effect of BMI and WHR (ANCOVA). All analysis was done for the whole group, but some data are presented for obese and nonobese separately. All analysis was performed using SPSS and the R software package.

| Characteristics                  | PCOS (n = 114) | Controls (n = 41) |
|----------------------------------|----------------|------------------|
| AGE (years)                      | 24.88 ± 0.59   | 26.60 ± 0.89     |
| BMI (kg/m²)                      | 28.80 ± 0.67   | 28.55 ± 1.35     |
| WHR                              | 0.81 ± 0.006   | 0.82 ± 0.015     |
| Fasting plasma glucose (mmol/L)  | 4.42 ± 0.06    | 4.37 ± 0.09      |
| Total testosterone (nmol/L)      | 3.73 ± 0.16    | 1.89 ± 0.10      |
| Systolic BP (mmHg)               | 125.12 ± 1.13  | 126.39 ± 2.30    |
| Diastolic BP (mmHg)              | 80.61 ± 0.93   | 82.92 ± 1.85     |

*p < 0.05.
Table 2: Investigated indices in PCOS and controls.

| Parameter                                      | PCOS         | Controls     | P value |
|------------------------------------------------|--------------|--------------|---------|
| Fasting glucose (mmol/L)                       | 4.42 ± 0.06  | 4.37 ± 0.089 | p > 0.05|
| Plasma glucose at 120 min of OGTT (mmol/L)     | 5.29 ± 0.10  | 4.72 ± 0.17  | p < 0.05|
| Fasting insulin (mU/L)                         | 16.35 ± 0.99 | 12.34 ± 1.47 | p < 0.05|
| Plasma insulin at 120 min of OGTT (mU/L)       | 78.52 ± 5.93 | 52.07 ± 8.69 | p < 0.05|
| Area under glucose curve (OGTT)                | 729.47 ± 13.43 | 699.40 ± 24.81 | p > 0.05|
| Area under insulin curve (OGTT)                | 9931.62 ± 594.49 | 7816.08 ± 892.38 | p < 0.05|
| Si (insulin sensitivity, minimal model analysis)| 2.49 ± 0.18  | 3.41 ± 0.36  | p < 0.05|
| AIR (acute insulin response, minimal model analysis) | 76.29 ± 4.56 | 65.69 ± 3.28 | p > 0.05|
| Di (disposition index, minimal model analysis) | 171.07 ± 13.07 | 220.28 ± 28.12 | p > 0.05|

Figure 1: Plasma glucose at 2 hr of OGTT in PCOS patients and controls. PCOS vs. controls, p < 0.05. Data are presented as the mean ± SEM, separately for nonobese subjects (BMI < 25 kg/m²) and overweight/obese (BMI > 25 kg/m²) (a). Relationship between BMI and plasma glucose at 2 hr of OGTT (b). Relationship between WHR and plasma glucose at 2 hr of OGTT (c). Relationship between age and plasma glucose at 2 hr of OGTT (d). *p < 0.05.
Table 3: Investigated indices in PCOS and controls (nonobese vs. overweight/obese).

| Parameter                                      | PCOS vs. controls (nonobese) | P value | PCOS vs. controls (overweight/obese) | P value |
|------------------------------------------------|------------------------------|---------|--------------------------------------|---------|
| Fasting glucose (mmol/L)                       | 4.34 ± 0.11 vs. 4.14 ± 0.13 | p > 0.05| 4.46 ± 0.08 vs. 4.58 ± 0.10          | p > 0.05|
| Plasma glucose at 120 min of OGTT (mmol/L)     | 4.87 ± 0.16 vs. 4.26 ± 0.23 | p < 0.05| 5.50 ± 0.13 vs. 5.10 ± 0.21          | p > 0.05|
| Fasting insulin (mU/L)                         | 12.33 ± 1.35 vs. 8.93 ± 1.09| p > 0.05| 18.35 ± 2.8 vs. 15.58 ± 2.51         | p > 0.05|
| Plasma insulin at 120 min of OGTT (mU/L)       | 48.19 ± 5.13 vs. 31.50 ± 4.29| p < 0.05| 93.68 ± 7.99 vs. 71.65 ± 15.45       | p < 0.05|
| Area under glucose curve (OGTT)                | 669.28 ± 21.58 vs. 651.53 ± 37.89| p < 0.05| 759.57 ± 16.01 vs. 744.99 ± 29.86    | p < 0.05|
| Area under insulin curve (OGTT)                | 7163.16 ± 252.17 vs. 5681.55 ± 460.25| p > 0.05| 11315.86 ± 736.86 vs. 9848.96 ± 1579.33| p < 0.05|
| Si (insulin sensitivity, minimal model analysis)| 3.39 ± 0.38 vs. 4.48 ± 0.52 | p > 0.05| 2.03 ± 0.39 vs. 2.40 ± 0.39          | p < 0.05|
| AIR (acute insulin response, minimal model analysis) | 61.78 ± 6.12 vs. 69.37 ± 4.47 | p > 0.05| 83.56 ± 5.96 vs. 62.21 ± 4.76        | p > 0.05|
| Di (disposition index, minimal model analysis)  | 190.29 ± 24.16 vs. 298.05 ± 45.58 | p < 0.05| 161.45 ± 15.44 vs. 146.22 ± 25.40    | p > 0.05|

![Figure 2: Basal plasma insulin in PCOS patients and controls.](image)

FIGURE 2: Basal plasma insulin in PCOS patients and controls. PCOS vs. controls, p < 0.05. Data are presented as mean ± SEM, separately for nonobese subjects (BMI < 25 kg/m²) and overweight/obese (BMI > 25 kg/m²) (a). Relationship between BMI and PCOS on basal plasma insulin (p < 0.001) (b). Relationship between WHR and PCOS on basal plasma insulin (p < 0.05) (c). *p < 0.05, **p < 0.001.
3. Results

3.1. Clinical Characteristics. The clinical characteristics of PCOS women and controls are presented in Table 1. There is no difference in age, BMI, WHR, fasting glucose, or blood pressure between PCOS and controls. In the PCOS group, 67.53% patients were overweight/obese. In control group 51.22% women were overweight/obese. Total testosterone levels were substantially higher ($p < 0.001$) in PCOS than in control women ($3.73 \pm 0.16$ vs. $1.89 \pm 0.10$ nmol/L). As regards PCOS clinical phenotypes, 109 (95.62%) women had hyperandrogenism and polycystic ovary morphology (PCOM) and 4.38% (5 women) had hyperandrogenism and oligoovulation.

3.2. Fasting Plasma Glucose, Insulin, and OGTT. After adjusting for BMI and using an analysis of covariance, it was observed that there was no difference in fasting plasma glucose between PCOS and controls ($4.42 \pm 0.06$ vs. $4.37 \pm 0.09$ mmol/L, $p > 0.05$) (Table 2). There was a positive interaction between BMI and PCOS ($p < 0.05$) (Figure 1(a), Table 2) and in nonobese subgroup of PCOS in comparison with nonobese control (Table 3). BMI (Figure 1(b)) and WHR (Figure 1(c)) were positively correlated with plasma glucose at 2 hr of OGTT while positively correlated with this parameter only in the control group (Figure 1(d)). AUCG was not significantly higher in PCOS group vs. controls ($729.47 \pm 13.43$ vs. $699.40 \pm 24.81$ mmol/L/120 min, $p > 0.05$ (Table 2)).

Fasting plasma insulin and plasma insulin at 2 hr of OGTT were significantly higher in PCOS patients than in controls ($p < 0.05$) (Figures 2(a) and 3(a)) while AUCI was not
significantly higher in PCOS group vs. controls (9931.62 ± 594.49 vs. 7816.08 ± 892.38 mU/L/120 min, p > 0.05) (Tables 2 and 3). BMI and WHR correlated positively with these parameters (p < 0.05) (Figures 2(b) and 3(b)). Significant interactions were found between PCOS and BMI on basal and stimulated plasma insulin (p < 0.001) (Figures 2(b) and 3(b)), as well as between WHR and PCOS on basal/stimulated insulin (p < 0.001, respectively) (Figures 2(c) and 3(c)).

3.3. Minimal Model Assessment of $S_i$ and $AIR_G$. $S_i$ was decreased in patients with PCOS compared to controls (p < 0.05) (Figure 4(a)). There was a significant interaction between PCOS and BMI as well as between WHR and PCOS in relation to $S_i$ (p < 0.001) (Figures 4(b) and 4(c)). Results obtained by measuring $AIR_G$ show that $AIR_G$ was not different between the PCOS and the control group (p > 0.05) (Figure 5(a)). With increasing age, $AIR_G$ decreased in both groups. Interaction between age and PCOS on this parameter shows that $AIR_G$ declines more with aging in the PCOS group (p < 0.001) (Figure 5(b)). Disposition index ($S_i \times$ AIR) was decreased in the PCOS group but not significantly compared to controls (p > 0.05) (Figure 6). When PCOS patients and controls were collated into two subgroups based on age (subgroup A < 25 years, subgroup B ≥ 25 years) $AIR_G$ was higher in the PCOS subgroup A than in age-matched controls (p < 0.05) (Figure 7(a)). $S_i$ was decreased in the PCOS group (p < 0.05) and DI was not significantly decreased in the PCOS subgroup A compared to the control subgroup A (p = 0.061) (Figures 7(b) and 7(c)) (Table 4). The entire group of women with PCOS had a
normal glucose tolerance during IVGTT (Kg), not different from controls (2.01 ± 0.11 vs. 1.88 ± 0.14 × 10², p > 0.05). A positive relationship between BMI and WHR with PCOS was also confirmed (p < 0.05). The best predictors of BMI as well as WHR were presented in Table 5.

4. Discussion

The prevalence of impaired glucose tolerance (IGT) and diabetes increased in PCOS [14, 15]. Our data confirmed that plasma glucose at the 2 hr point of OGTT was higher in the PCOS group than in controls although all investigated subjects had normal fasting glucose and normal glucose tolerance. The PCOS subjects in this investigation had higher basal plasma insulin and higher plasma insulin levels at 2 hr of OGTT compared to controls. Our results agree with the notion that high BMI and central obesity cause exaggerated insulin responses in PCOS women [16, 17]. Insulin resistance occurs in 40%–70% of women with PCOS [18]. Obesity may increase PCOS prevalence and exacerbate IR in women with PCOS [19], while insulin resistance in lean women with PCOS is not consistently demonstrated [20]. The euglycemic-hyperinsulinemic clamp is the gold standard to directly measure insulin sensitivity. In some previous studies [21, 22] it was showed that women with PCOS have intrinsic reduction in insulin sensitivity on euglycemic-hyperinsulinemic clamp and almost all obese women with PCOS have more serious IR than lean women with PCOS. A systematic review and meta-analysis of euglycemic-hyperinsulinemic clamp studies by Cassar et al. showed a reduction in insulin sensitivity of 27% and obesity exacerbates the reduction in insulin sensitivity by 15% in women with PCOS [23]. Our results confirmed the existence of insulin resistance in young PCOS patients. After adjusting for BMI, S_i remained lower in PCOS women than in controls. Overweight contributed to the impairment of insulin sensitivity in PCOS as well as controls [24]. Furthermore, our results showed that the interaction between disease (PCOS) and being overweight exists. These data suggest that increased BMI may have a more deleterious effect on insulin sensitivity in PCOS than in controls. This is important because overweight or obesity is detected in about 30–50% women with PCOS [9]. Furthermore, our data indicate that individual lean patients with PCOS often have no hyperinsulinemia and insulin resistance as it was shown in previous study [25].

Android obesity, clinically confirmed as increased WHR, is an especially strong risk for insulin resistance and other factors that predispose to premature cardiovascular disease [20]. Although we were unable to demonstrate a difference in WHR between PCOS and healthy women, our results
Table 4: Investigated indices in PCOS and controls (<25 years old vs. ≥25 years old).

| Parameter                              | PCOS vs. controls (<25 years old) | P value | PCOS vs. controls (≥25 years old) | P value |
|----------------------------------------|-----------------------------------|---------|-----------------------------------|---------|
| Fasting glucose (mmol/L)               | 4.39 ± 0.09 vs. 4.33 ± 0.18       | p < 0.05| 4.45 ± 0.09 vs. 4.39 ± 0.09       | p < 0.05|
| Plasma glucose at 120 min of OGTT (mmol/L) | 5.16 ± 0.14 vs. 4.36 ± 0.23 | p < 0.05| 5.44 ± 0.15 vs. 4.94 ± 0.24 | p < 0.05|
| Fasting insulin (mU/L)                 | 16.09 ± 1.18 vs. 14.02 ± 3.20    | p < 0.05| 16.63 ± 1.66 vs. 11.26 ± 1.29    | p < 0.05|
| Plasma insulin at 120 min of OGTT (mU/L) | 86.30 ± 9.82 vs. 42.42 ± 6.49 | p < 0.05| 69.87 ± 6.04 vs. 58.24 ± 13.61 | p < 0.05|
| Area under glucose curve (OGTT)       | 704.59 ± 18.19 vs. 665.72 ± 32.78 | p < 0.05| 757.12 ± 19.36 vs. 720.96 ± 34.69 | p < 0.05|
| Area under insulin curve (OGTT)       | 10508.03 ± 343.95 vs. 700.97 ± 834.59 | p = 0.061| 9291.17 ± 703.45 vs. 8337.75 ± 1367.11 | p < 0.05|
| Si (insulin sensitivity, minimal model analysis) | 2.43 ± 0.25 vs. 4.52 ± 0.62 | p < 0.05| 2.54 ± 0.27 vs. 2.71 ± 0.38 | p < 0.05|
| AIR (acute insulin response, minimal model analysis) | 92.57 ± 6.88 vs. 64.14 ± 3.88 | p < 0.05| 58.22 ± 4.82 vs. 66.69 ± 4.82 | p < 0.05|
| Di (disposition index, minimal model analysis) | 195.77 ± 18.89 vs. 308.23 ± 53.15 | p > 0.05| 143.61 ± 17.34 vs. 163.99 ± 26.33 | p > 0.05|

Figure 7: AIR and in age subgroups of PCOS patients and controls. Subgroup A: <25 years old. Subgroup B: ≥25 years old. Data are presented as the mean ± SEM (a). Si in age subgroups of PCOS patients and controls. Data presented as the mean ± SEM (b). DI in age subgroups of PCOS patients and controls. Data presented as the mean ± SEM (c).
confirmed a significant influence of android obesity towards insulin resistance in both PCOS and controls. Thus, our data also indicated that PCOS women are more susceptible to increasing WHR regarding the development of insulin resistance.

The product of insulin sensitivity (Si) and insulin response (\(AIRE_\beta\)) in healthy subjects is a constant value that has been termed the disposition index (DI) [12]. It is expected that \(AIRE_\beta\) will be higher as a compensatory mechanism in the state of insulin resistance [13]. On the other hand, the DI is low in subjects with IGT or T2DM [25]. In our study, we demonstrated decreased DI in nonobese PCOS compared to controls. A possible explanation for this finding is the lack of a compensatory insulin response in nonobese PCOS women as would be expected in an insulin resistant state.

Interestingly, the data show that there is no difference in \(AIRE_\beta\) between PCOS and controls. Results from this study clearly show a significant negative correlation between \(AIRE_\beta\) and increasing age. With an increase in age, \(AIRE_\beta\) falls faster in patients with PCOS. Interestingly, our data did not confirm the significant influence of obesity on this parameter [24].

While insulin resistance is a factor in the development of PCOS, the associated failure of pancreatic \(\beta\)-cell function could be an important determinant of the development of T2DM in many of these women [26–28]. Although most of the observed hyperinsulinemia in PCOS is probably secondary to insulin resistance, there seems to be an important component of abnormal insulin secretion, which is independent of insulin resistance, body weight, and body fat distribution [3, 8, 29, 30]. Thus, besides insulin resistance, \(\beta\)-cell dysfunction seems to be an integral characteristic of this syndrome [27]. A recent study showed that in women with PCOS, metabolic clearance of insulin is reduced, contributing to developing hyperinsulinemia, as well as that serum androgens are independent predictors of this phenomenon [28]. But it was demonstrated that a higher prevalence of impaired insulin secretion than impaired insulin action exists in first-degree relatives of patients with PCOS [27, 31].

According to a previously published report [26], \(\beta\)-cell secretory defects may contribute to increased carbohydrate intake and subsequently obesity and insulin resistance. Current data support the notion that glucose intolerance and frank T2DM are found in combination with the sign of \(\beta\)-cell exhaustion in a significant portion of young women with PCOS. These subjects with PCOS may manifest later stage carbohydrate intolerance and T2DM after long-standing insulin resistance in susceptible women [26]. In this regard, our data could suggest that an intrinsic defect in insulin secretion exists in PCOS patients.

In the youngest PCOS women (subgroup A), we demonstrated exaggerated \(AIRE_\beta\) in comparison with age-matched controls as well as compared to older PCOS women (subgroup B). The exaggerated \(AIRE_\beta\) in our young patients with PCOS could be a compensatory response to underlying insulin resistance. Despite the compensatory response, this group failed to achieve a DI observed in controls.

Current data are in concert with prior reports that significant abnormalities in insulin secretion are already present in patients with PCOS <25 years [31, 32]. Although, in our younger PCOS women, \(AIRE_\beta\) was enhanced, the DI was still decreased compared to age-matched controls due to significantly impaired \(S_i\), placing these patients at heightened risk for T2DM. Studies on young adolescent girls may provide clues about the pathophysiology of PCOS since this is an age when early clinical signs are manifested. Thus, this may be the time to initiate hygienic and medical interventions to retard the development of impaired glucose intolerance and T2DM [27, 29–31, 33]. This study suggests that young PCOS women can indeed be identified and placed on therapy to reduce the cardiovascular risk factors and development of T2DM [10, 34–49].

### 5. Conclusion

The major strength of this study is the large number of PCOS women investigated using minimal model analyses to evaluate acute insulin response and assess insulin secretion as well as insulin sensitivity. However, there are also limitations in the study. This is an observational study, and cause-effect relationships cannot be firmly established. In addition, as most women in this cohort had a hyperandrogenic PCOS phenotype, the results of the study may be more applicable to these subjects.

In conclusion, current observations underline the importance of interactions between PCOS, BMI, age, and WHR. This investigation has also shown that not all patients with PCOS demonstrate basal and stimulated hyperinsulinemia, insulin resistance, and impaired glucose tolerance, particularly early in the evolution of PCOS as a clinical entity. Our data concerning subjects younger than 25 years underscores the importance of establishing the diagnosis of PCOS in adolescence, and the institution of appropriate therapy targeting insulin resistance and \(\beta\)-cell secretion before T2DM develops.

### Data Availability

Data can be made available on reasonable request (msumaracdumanovic@gmail.com).

---

**Table 5: Regression analysis: best predictors of BMI and WHR.**

|      | Estimate | Std. error | Coefficient | p value |
|------|----------|------------|-------------|---------|
| BMI  | Intercept| 24.12239   | 2.48639     |         |
|     | Age      | 0.20362    | 0.08492     |         |
|     | Basal insulin | 0.19604 | 0.05077     |         |
|     | Si       | −1.29620   | 0.25430     |         |

|      | Estimate | Std. error | Coefficient | p value |
|------|----------|------------|-------------|---------|
| WHR  | Intercept| 0.67519190 | 0.03245457  | 20.804  |
|     | Age      | 0.00352968 | 0.00088745  | 3.977   |
|     | AUCG     | 0.0009734  | 0.0003786   | 2.571   |
|     | Si       | −0.00778025 | 0.00255817  | −3.041  |

\[ r = 0.521 \quad < 0.001 \]

\[ r = 0.474 \quad < 0.001 \]
Conflicts of Interest

The authors declare that there are no conflicts of interest regarding the publication of this paper.

Acknowledgments

The research and publication of this article were funded by the School of Medicine and Clinical Center of Serbia.

References

[1] S. Franks, “Polycystic ovary syndrome,” New England Journal of Medicine, vol. 333, no. 13, pp. 853–861, 1995.
[2] The Rotterdam ESHRE/ASRM-Sponsored PCOS Consensus Workshop Group, “Revised 2003 consensus on diagnostic criteria and long-term health risks related to polycystic ovary syndrome (PCOS),” Human Reproduction, vol. 19, pp. 41–47, 2004.
[3] E. B. Johnstone, M. P. Rosen, R. Neril et al., “The polycystic ovary post-Rotterdam: a common, age-dependent finding in ovulatory women without metabolic significance,” Journal of Clinical Endocrinology & Metabolism, vol. 95, no. 11, pp. 4965–4972, 2010.
[4] J. E. Nestler, “Role of hyperinsulinemia in the pathogenesis of the polycystic ovary syndrome, and its clinical implications,” in Seminars in Reproductive Endocrinology, pp. 111–122, Thieme Medical Publishers Inc, New York, NY, USA, 1997.
[5] N. M. O’Meara, J. D. Blackman, D. A. Ehrmann et al., “Defects in beta-cell function in functional ovarian hyperandrogenism,” Journal of Clinical Endocrinology & Metabolism, vol. 76, no. 5, pp. 1241–1247, 1993.
[6] R. S. Legro, A. R. Kunselman, W. C. Dodson, and A. Dunaif, “Prevalence and predictors of risk for type 2 diabetes mellitus and impaired glucose tolerance in polycystic ovary syndrome: a prospective, controlled study in 254 affected women,” Journal of Clinical Endocrinology & Metabolism, vol. 84, no. 1, pp. 165–169, 1999.
[7] H. Bili, J. Laven, B. Imani, M. J. Eijkemans, and B. C. Fauser, “Age-related differences in features associated with polycystic ovary syndrome in normogonadotrophic oligo-amenorrheic infertile women of reproductive years,” European Journal of Endocrinology, vol. 145, pp. 749–755, 2001.
[8] S. A. Arslanian, V. D. Lewy, and K. Danadian, “Glucose intolerance in obese adolescents with polycystic ovary syndrome: roles of insulin resistance and β-cell dysfunction and risk of cardiovascular disease,” Journal of Clinical Endocrinology & Metabolism, vol. 86, no. 1, pp. 66–71, 2001.
[9] E. Diamanti-Kandarakis and A. Dunaif, “Insulin resistance and the polycystic ovary syndrome revisited: an update on mechanisms and implications,” Endocrine Reviews, vol. 33, no. 6, pp. 981–1030, 2012.
[10] World Health Organization Study Group, “Diabetes mellitus,” World Health Organization Technical Report Series, vol. 727, pp. 1–113, 1985.
[11] R. N. Bergman, Y. Z. Ider, C. R. Bowden, and C. Cobelli, “Quantitative estimation of insulin sensitivity,” American Journal of Physiology, Endocrinology and Metabolism, vol. 236, no. 6, Article ID E667, 1979.
[12] G. Pacini and R. N. Bergman, “MINMOD: a computer program to calculate insulin sensitivity and pancreatic responsivity from the frequently sampled intravenous glucose tolerance test,” Computer Methods and Programs in Biomedicine, vol. 23, no. 2, pp. 113–122, 1986.
[13] R. N. Bergman, D. T. Finegood, and S. E. Kahn, “The evolution of β-cell dysfunction and insulin resistance in type 2 diabetes,” European Journal of Clinical Investigation, vol. 32, pp. 35–45, 2002.
[14] A. Miller, Leaps: Regression Subset Selection. R package version 3.1, 2000, https://CRAN.R-project.org/package=leaps
[15] J. J. Conn, H. S. Jacobs, and G. S. Conway, “The prevalence of polycystic ovaries in women with type 2 diabetes mellitus,” Clinical Endocrinology, vol. 52, no. 1, pp. 81–86, 2000.
[16] R. Pasquali, F. Casimirri, S. Venturoli et al., “Body fat distribution has weight-independent effects on clinical, hormonal, and metabolic features of women with polycystic ovary syndrome,” Metabolism, vol. 43, no. 6, pp. 706–713, 1994.
[17] F. Ovalle and R. Azziz, “Insulin resistance, polycystic ovary syndrome, and type 2 diabetes mellitus,” Fertility and Sterility, vol. 77, no. 6, pp. 1095–1105, 2002.
[18] C. M. De Ugarte, A. Bartolucci, and A. Azziz, “Prevalence of insulin resistance in the polycystic ovary syndrome using the homeostasis model assessment,” Fertility and Sterility, vol. 83, no. 5, pp. 1454–1460, 2005.
[19] H. Teede, A. Deeks, and L. Moran, “Polycystic ovary syndrome: a complex condition with psychological, reproductive and metabolic manifestations that impacts on health across the lifespan,” BMC Medicine, vol. 8, no. 1, 41 pages, 2010.
[20] G. Gennarelli, V. Roveri, R. F. Novi et al., “Preserved insulin sensitivity and β-cell activity, but decreased glucose effectiveness in normal-weight women with the polycystic ovary syndrome,” Journal of Clinical Endocrinology & Metabolism, vol. 90, no. 6, pp. 3381–3386, 2005.
[21] N. K. Stepto, S. Cassar, A. E. Joham et al., “Women with polycystic ovary syndrome have intrinsic insulin resistance on euglycaemic-hyperinsulinaemic clamp,” Human Reproduction, vol. 28, no. 3, pp. 777–784, 2013.
[22] F. Tosi, E. Bonora, and P. Moghetti, “Insulin resistance in a large cohort of women with polycystic ovary syndrome: a comparison between euglycaemic-hyperinsulinaemic clamp and surrogate indexes,” Human Reproduction, vol. 32, no. 12, pp. 2515–2521, 2017.
[23] S. Cassar, M. L. Misso, W. G. Hopkins, C. S. Shaw, H. J. Teede, and N. K. Stepto, “Insulin resistance in polycystic ovary syndrome: a systematic review and meta-analysis of euglycaemic-hyperinsulinaemic clamp studies,” Human Reproduction, vol. 31, pp. 2619–2631, 2016.
[24] A. Dunaif and D. T. Finegood, “Beta-cell dysfunction independent of obesity and glucose intolerance in the polycystic ovary syndrome,” Journal of Clinical Endocrinology & Metabolism, vol. 81, no. 3, pp. 942–947, 1996.
[25] P. Ovesen, J. E. N. S. Moller, H. J. Ingerslev et al., “Normal basal and insulin-stimulated fuel metabolism in lean women with the polycystic ovary syndrome,” Journal of Clinical Endocrinology & Metabolism, vol. 77, no. 6, pp. 1636–1640, 1993.
[26] J. Holte, T. Bergh, C. Berne, L. Berghlund, and H. Lithell, “Enhanced early insulin response to glucose in relation to insulin resistance in women with polycystic ovary syndrome and normal glucose tolerance,” Journal of Clinical Endocrinology & Metabolism, vol. 78, no. 5, pp. 1052–1058, 1994.
[27] M. K. Cavaghan, D. A. Ehrmann, and K. S. Polonsky, “Interactions between insulin resistance and insulin secretion in the development of glucose intolerance,” Journal of Clinical Investigation, vol. 106, no. 3, pp. 329–333, 2000.
[28] F. Tosi, F. Dal Molin, F. Zamboni et al., “Serum androgens are independent predictors of insulin clearance but not of insulin secretion in women with pcos,” *Journal of Clinical Endocrinology & Metabolism*, vol. 105, no. 5, p. e1981–e1989, 2020.

[29] J. Holte, T. Bergh, C. H. Berne, L. Wide, and H. Lithell, “Restored insulin sensitivity but persistently increased early insulin secretion after weight loss in obese women with polycystic ovary syndrome,” *Journal of Clinical Endocrinology & Metabolism*, vol. 80, no. 9, pp. 2586–2593, 1995.

[30] J. Holte, “Disturbances in insulin secretion and sensitivity in women with the polycystic ovary syndrome,” *Baillière's Clinical Endocrinology and Metabolism*, vol. 10, no. 2, pp. 221–247, 1996.

[31] S. Colilla, N. J. Cox, and D. A. Ehrmann, “Heritability of insulin secretion and insulin action in women with polycystic ovary syndrome and their first-degree relatives,” *Journal of Clinical Endocrinology & Metabolism*, vol. 86, no. 5, pp. 1555–1559, 2002.

[32] R. Homburg, “Should patients with polycystic ovarian syndrome be treated with metformin? A note of cautious optimism,” *Human Reproduction*, vol. 17, no. 4, pp. 853–856, 2002.

[33] J. C. Marshall and A. Dunaif, “All women with PCOS should be treated for insulin resistance,” *Fertility and Sterility*, vol. 97, no. 1, pp. 18–22, 2012.

[34] S. I. McFarlane, M. Banerji, and J. R. Sowers, “Insulin resistance and cardiovascular Disease,” *Journal of Clinical Endocrinology & Metabolism*, vol. 86, no. 2, pp. 713–718, 2001.

[35] D. H. Abbott, D. A. Dumesic, and S. Franks, “Developmental origin of polycystic ovary syndrome-a hypothesis,” *Journal of Endocrinology*, vol. 174, no. 1, pp. 1–5, 2002.