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ORIGINAL ARTICLE

Changes in insulin resistance, glucose effectiveness, and first and second phases of insulin secretion in women aged 45–60 years old in Taiwan

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Abstract

Background: In women after menopause, the incidence of diabetes mellitus increases. Increased insulin resistance (IR), decreased glucose effectiveness (GE), and the first and second phases of insulin secretion (FPIS and SPIS), are the four most important factors that trigger glucose intolerance and diabetes (diabetogenic factor [DF]). In the cross-sectional study, we enrolled nondiabetic women between the ages of 45 and 60 years to observe the changes in DFs during the perimenopausal period and to elucidate the underlying mechanisms of diabetes in menopausal women.

Methods: We randomly enrolled 4194 women who underwent health checkups. Using demographic and biochemical data, IR, FPIS, SPIS, and GE were calculated using previously published equations. The relationship between the DFs and age was evaluated using a simple correlation.

Results: Body mass index, blood pressure, fasting plasma glucose, low-density lipoprotein cholesterol, triglyceride, and SPIS were higher, and GE was lower in older women (≥52 years old). A significant decrease in GE and increased SPIS were observed with age. However, no changes were observed in IR or FPIS.

Conclusion: The IR and FPIS did not change during perimenopause. Increased SPIS may compensate for the decrease in GE, which is probably one of the reasons for the higher incidence of diabetes in menopausal women.

Keywords: Insulin resistance; Insulin secretion; Perimenopause

1. INTRODUCTION

Type 2 diabetes (T2D) is one of the five main causes of death in Taiwan and many other countries,[1,2] burdening affected individuals, their families, and society.[3,4] Therefore, prevention and early detection of the disease is a public health priority. The correlation between an increased incidence of T2D and postmenopausal women has long been studied. The findings of most cross-sectional or longitudinal studies on the correlation between natural menopause and the risk of diabetes are controversial.[5–9] However, the results differed in women with surgically induced menopause. The aforementioned studies unanimously indicated a significantly higher risk of developing diabetes in surgically induced menopausal women.[10–13] The potential mechanisms linking menopause and diabetes include increased adiposity and changes in sex hormones.[14–17] The most commonly investigated underlying pathophysiologies of T2D are insulin resistance (IR) and impaired insulin secretion.[18] However, evidence suggests that glucose effectiveness (GE) is important in glucose intolerance.[19] In addition, there are two phases of insulin secretion: the first and second phases (FPIS and SPIS, respectively).[20,21] Due to the complexity of measuring the four diabetogenic factors (DF: IR, GE, FPIS, and SPIS), their roles in menopause are poorly understood. In this cross-sectional study, we enrolled healthy women aged 45 to 60 years without a history of diabetes or any related medications to observe the changes in DFs. Therefore, the study aimed to elucidate the underlying mechanisms and effects of menopause on the risk of developing diabetes.

2. METHODS

2.1. Study participants

We randomly enrolled 4194 women aged 45 to 60 years old from Cardinal Tien Hospital and Tri-service General Hospital.

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Fig. 1 illustrates the process. The age bracket of 45 to 60 years was based on the average menopausal age of Taiwanese women, 52 years.[22] We included women 7 and 8 years older than the average to observe the changes in DFs. All participants were anonymous, and they provided written informed consent. The institutional review boards of Cardinal Tien Hospital and Tri-service General Hospital approved the study protocols (TSGH-100-05-246 and CTH-100-2-5-036, respectively). Participants who were obese (body mass index [BMI] ≥25 kg/m^2), patients with diabetes (fasting plasma glucose [FPG] ≥126), hypertension and/or hyperlipidemia, or on any medications known to affect blood pressure, glucose, and lipids levels were excluded. On the day of the study, senior nursing staff recorded participants' medical histories, including information on any current medications. A physical examination was also performed. Moreover, waist circumference (WC) was measured horizontally at the level of the natural waist, whereas BMI was calculated as the participant's body weight (kg) divided by the square of the participant's height (m). Systolic blood pressure (SBP) and diastolic blood pressure (DBP) were measured using standard mercury sphygmomanometers placed on the right arm of each seated participant. After fasting for 10 hours, blood samples were collected for biochemical analyses. Plasma was separated from the blood within 1 hour and stored at 30°C until further analysis of FPG and lipid profiles. The glucose oxidase method was used to measure FPG (YSI 203 glucose analyzer; Yellow Springs Instruments, Yellow Springs, OH, USA). Total cholesterol and triglycerides (TG) were measured using the dry multilayer analytical slide method with a Fuji Dri-Chem 3000 analyzer (Fuji Photo Film, Tokyo, Japan). Serum high-density lipoprotein cholesterol (HDL-C) and low-density lipoprotein cholesterol (LDL-C) concentrations were analyzed using an enzymatic cholesterol assay following dextran sulfate precipitation. We calculated the IR, FPIS, SPIS, and GE using the following equations; all the units are international units. For gender, "1" denotes men, and "2" denotes women:

The equations used to calculate the IR, FPIS, SPIS, and GE are as follows: To demonstrate the reliability of our equations, we provided the following short statement. Approximately 70% of the participants were used to build the equation, and the remaining 30% were used for external validation. Therefore, the accuracies of the equations were tested. The equations used in the study have been previously published. The conclusions are as follows:

1. IR: The study enrolled 327 participants and the IR was measured using an insulin suppression test. The *r* value between the measured and calculated GE values was 0.581 ($p < 0.001$). The equation was first published in 2013 in the *Journal of Diabetes Investigation*. IR = log (1.439 + 0.018 × sex – 0.003 × age + 0.029 × BMI – 0.001 × SBP + 0.006 × DBP + 0.049 × TG – 0.046 × HDL-C – 0.0116 × FPG) × $10^{3.333,23}$
2. FPIS: The study enrolled 186 participants. Moreover, FPIS was measured using a frequently sampled intravenous glucose tolerance test. The *r* value between the measured and calculated GE values was 0.671 ($p < 0.000$). The equation was first published in the *International Journal of Endocrinology* in 2015.

FPIS = $10^{(1.477 - 0.119 \times FPG + 0.079 \times BMI - 0.523 \times HDL\text{-}C)24}$

3. SPIS: The study enrolled 82 participants. SPIS was measured using a modified low-dose glucose infusion test. The *r* value between the measured and calculated GE was 0.65 ($p = 0.002$). The equation was published in the *Metabolic Syndrome and Related Disorders* journal in 2016.

SPIS = $10^{(-2.4 - 0.088 \times FPG + 0.072 \times BMI)25}$

GE: The study enrolled 227 participants. The GE levels were measured using a frequently sampled intravenous glucose

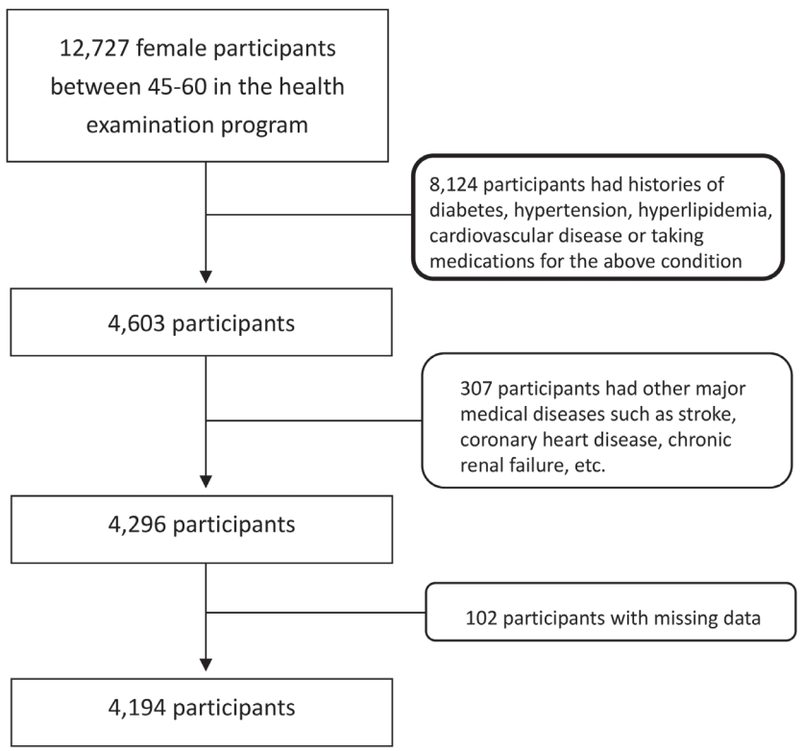

Fig. 1 The exclusion and inclusion flow of the study participants.

tolerance test. The *r* value between the measured and calculated GE was 0.43 ($p = 0.001$). The following equation was published in the *Metabolic Syndrome and Related Disorders* journal in 2016.

$$GE = (29.196 - 0.103 \times age - 2.722 \times TG - 0.592 \times FPG) \times 10^{-3}$$ [3,26]

2.2. Statistical analysis

All the statistical analyses were performed using SPSS 19.0 (IBM Inc., Armonk, New York, USA). Data are presented as mean ± SD. All data were tested for normal distribution using the Kolmogorov–Smirnov test and for homogeneity of variances using Levene's test. Data were log-transformed before analysis as the FPIS and SPIS were not normally distributed. A *t* test was performed to evaluate the differences between younger and older women, with a cutoff point of 52 years (older: ≥52 years old). A simple correlation was used to evaluate the relationship between age and DFs.

3. RESULTS

Table 1 displays the demographic, biochemical, and DFs data of the younger and older women. As compared with younger women, BMI, BP, FPG, LDL-cholesterol, TG, and SPIS were significantly higher in older women. However, GE was lower in older women compared with younger ones. No changes were observed in the IR, FPIS, or HDL cholesterol levels. Table 1 demonstrates a negative correlation between GE and age ($r = -0.41$), and a positive correlation between age and SPIS ($r = 0.056$). Additionally, Table 2 presents the simple correlations between age and DFs. No significant relationship was observed between IR and FPIS. Fig. 2 displays the graphic illustrations regarding the changes in the four DFs means regarding age. Furthermore, only the changes in the GE and SPIS scores were statistically significant.

Table 1

The demographic data of study subjects

Female	
n	4149
Age	52 ± 5
Body mass index (kg/m^2)	23 ± 2
Systolic blood pressure (mmHg)	128 ± 19
Diastolic blood pressure (mmHg)	76 ± 11
Fasting plasma glucose (mmol/dL)	5.31 ± 0.47
HDL-C (mmol/dL)	1.32 ± 0.35
LDL-C (mmol/dL)	3.57 ± 0.87
Triglyceride (mmol/dL)	1.33 ± 0.66
GE (10^{-2}·dL·min^{1}·kg^{-1})	0.017 ± 0.02
IR (10"4·min"1·pmor1·L^{-1})	3.68 ± 0.02
FPIS (μU/min)	95.5 ± 1.9
SPIS (pmol/mmol)	0.063 ± 1.44

Data shown are mean ± SD.

GE = glucose effectiveness; HDL-C = high-density lipoprotein cholesterol; IR = insulin resistance; LDL-C = low-density lipoprotein cholesterol; Log FPIS = log transformation of first-phase insulin secretion; Log SPIS = log transformation of second-phase insulin secretion.

Table 2

The results of Pearson correlation between age and glucose effectiveness, insulin resistance, first- and second-phase insulin secretion

	r	*p*
Female		--
Glucose effectiveness	–0.410	<0.001
Insulin resistance	–0.025	0.109
Log transformation of first-phase insulin secretion	0.020	0.185
Log transformation of second-phase insulin secretion	0.056	<0.001

4. DISCUSSION

The study demonstrated simultaneous changes in four important pathophysiological factors related to diabetes before and after menopause using estimation equations. Our results demonstrated that during the aforementioned period, there was a significant decrease in GE and an increase in SPIS. All participants were nondiabetic and were not receiving any medication related to metabolic syndrome. Therefore, the results presented here are more reliable than those of other studies on this topic without excluding participants with these confounding factors. Our data are novel and provide insight into the factors influencing blood glucose levels in perimenopausal women. Several studies on the prevalence of diabetes in perimenopausal women can be grouped into three categories: cross-sectional studies, typically, these studies have large "n" numbers.[5–7,27,28] The second category is longitudinal studies, which are more persuasive.[8,10,29–32] Unfortunately, the results of the studies are inconsistent. The third category of study obtained evidence from observations of surgical menopause. Estradiol (E2) levels drop more abruptly in women with surgically induced menopause than in those with natural menopause. This allows researchers to evaluate the effect of E2 on the risk of developing diabetes with greater precision.[10,11] The studies demonstrate a significant increase in the risk of diabetes in surgically treated menopausal women. In summary, studies investigating the prevalence of diabetes and menopause are conflicting, except studies investigating surgically induced menopause. Moreover, we demonstrated that the IR and FPIS levels remained unchanged before and after menopause. Simultaneously, reduced GE and increased SPIS act in opposite directions to increase diabetes prevalence. Therefore, we concluded that the association between menopause and diabetes is relatively weak. The question regarding the effect of menopause on diabetes concerns the underlying mechanism. As mentioned in the Introduction section, decreased levels of insulin secretion and GE or an increase in IR could contribute to this effect. Moreover, except for surgical menopause, the increased incidence of diabetes after menopause is controversial. Therefore, we aimed to explore the possible pathophysiological factors that cause diabetes (IR, FPIS, SPIS, and GE). Regarding the relationships among the four DFs, neither IR nor FPIS changed and only GE and FPIS were altered. Although the actual roles of GE are unknown, islet cells are known to have very good resilience to any change in plasma glucose levels. Thus, we hypothesized that the reduction of GE increases plasma glucose and, indirectly, SPIS increases to correct the derangements in glucose homeostasis. The important roles of lipids, BP, or BMI in diabetes are well known. The variables were included in the equations used in this study. Similarly, we adjusted for the traditional factors in the comparison. In the following sections, we discuss our results about the factors mentioned above. Our results established that SPIS scores increased in women but not FPIS scores. This finding is inconsistent with the findings of other mainstream studies that reported a significant decrease in insulin secretion in older women. Several factors might contribute to the discrepancy, such as the use of different methods for the measurement of-cell function, observation of different cohorts, and whether the participants were receiving medication. However, GE should also be considered to explain the aforementioned finding. Notably, the GE deteriorated dramatically with age. We hypothesized that cells compensate for the

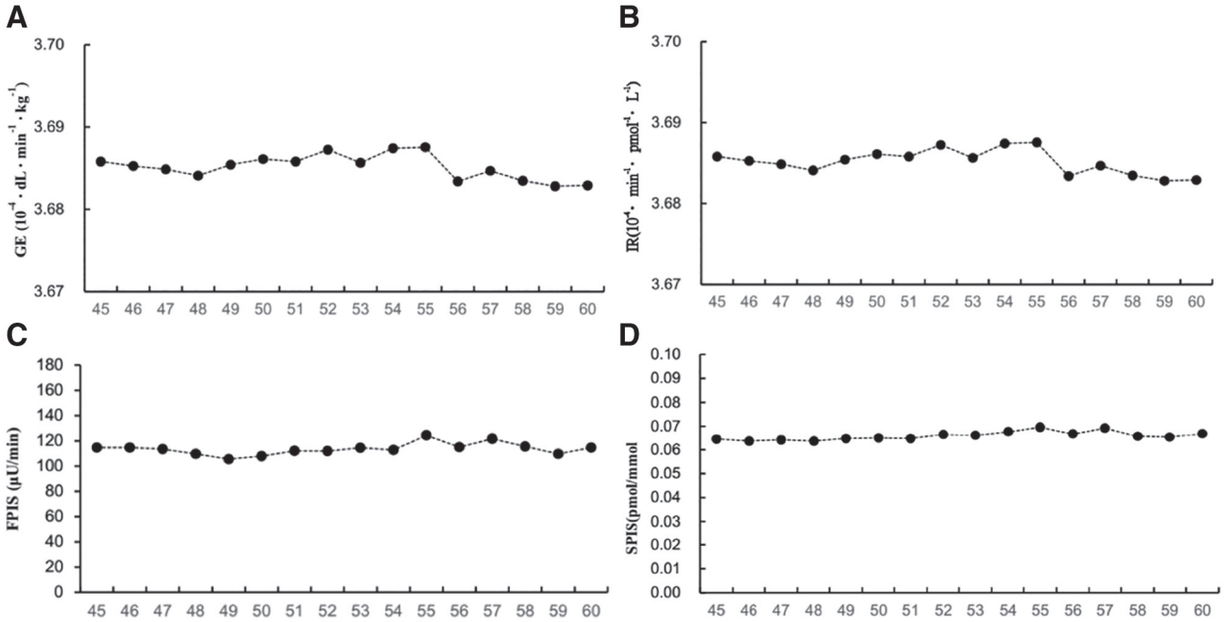

Fig. 2 The relationship between the participant's age and the mean of GE (A), IR (B), FPIS (C), and SPIS (D). FPIS = first-phase insulin secretion; GE = glucose effectiveness; IR = insulin resistance; FPIS = second-phase insulin secretion.

worsening of glucose tolerance. As the participants in the study were non-diabetic and in relatively good health, their ability to compensate was preserved. The reason FPIS did not increase simultaneously can be explained as follows. According to data from the United Kingdom Prospective Diabetes Study, FPIS disappeared 10 years before diabetes was diagnosed.[33] The observation suggests that, compared with the SPIS, the FPIS is more vulnerable to changes in other factors, such as increased IR and/or decreased GE. Thus, the fact that there was no change in FPIS is understandable. A recent report by Chandler et al[34] concluded that, although there have been contradictions in previous studies if BMI is adjusted, age does not affect IR levels.[34–37] In our study, the IR remained the same, which correlated with the findings of previous studies on changes in IR levels in women during menopause. Although evidence has exhibited that GE plays an important role in glucose metabolism, there are fewer studies on GE than on IR or insulin secretion,[19] one reason for this may be the difficulty of quantifying GE. As a result, little is known about the changes in GE during perimenopause. However, using a mixed-meal tolerance test with 6-(13)H glucose, Basu et al[36] identified no significant difference between the GE levels of older and younger women. In contrast, Burattini et al[38] demonstrated the opposite, establishing that age is an important predictor of GE. Additionally, it should be noted that both studies had very few "n" numbers, making their results less persuasive. Herein, we identified that the GE experienced the most profound deterioration during perimenopause, and as a result, SPIS increased to compensate. The finding explains the increased prevalence of diabetes after menopause.

The study has some limitations. First, as a cross-sectional study, the results are less persuasive than a longitudinal study. In the future, measuring DFs before and after menopause in the same participants could further validate our findings. Second, we did not have data on E2. If data for E2 levels were available, we could have extracted valuable information concerning the relationship between E2 and DFs and thus demonstrated the corresponding changes in their levels. Finally, one might argue that our methods for measuring the DFs are inaccurate. However, given that the methods were all validated by an intravenous glucose tolerance test[23–26] and that the changes in DFs could be explained physiologically, we are confident of the reliability of our methods.

In conclusion, we observed that the levels of IR and FPIS did not change in women aged 45 to 60 years old, mainly during the perimenopausal period. The increase in SPIS may compensate for the decrease in GE, which is likely one of the factors contributing to a higher incidence of diabetes in menopausal women. Therefore, more specific research is needed to uncover the mechanisms underlying the observed phenomenon.

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