Normal fasting plasma glucose predicts type 2 diabetes and cardiovascular disease in elderly population in Taiwan

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Summary

Background: Hyperglycemia increases prevalence of metabolic syndrome (MetS), type 2 diabetes (T2D) and cardiovascular disease (CVD). But the role of normoglycemia on the development of T2D and CVD in elderly population remains unclear.

Aim: To determine an optimal cut-off for fasting plasma glucose (FPG) to predict MetS and subsequent risk of T2D and CVD in an elderly Taiwanese population with normal FPG levels.

Design: Two stages included cross-sectional (Stage 1) and prospective (Stage 2) cohort study.

Methods: In Stage 1 18 287 subjects aged ≥60 years were enrolled; of these, 5039 without T2D and CVD advanced to Stage 2 and a mean follow-up of 3.8 years. MetS components were analysed, and in Stage 1, FPG cut-offs for MetS risk were calculated using receiver operating characteristic (ROC) curve analyses. In Stage 2, subjects without T2D and CVD in Stage 1 were classified into high-FPG and low-FPG groups based on cut-offs, and sex specific differences in incidence for T2D and CVD were calculated.

Results: ROC curve analysis gave an optimal FPG cut-off for MetS of 93 mg/dl and 92 mg/dl for males and females, respectively. The high-FPG group had a 1.599- and 1.353-fold higher chance of developing T2D compared with the low-FPG group for males and females, respectively (95% CI: 1.606–2.721 and 1.000–1.831, \( P = 0.015 \) and 0.05). The high-FPG group had a 1.24-fold higher chance of developing CVD for females (95% CI: 1.015–1.515, \( P = 0.035 \)); however, there was no difference for males.
Conclusions: Our results suggest that FPG within the normal range was associated with MetS, and elderly subjects with high normal levels have a higher incidence of developing T2D for both sexes, and CVD for females, over the short-term.

Introduction
Type 2 diabetes (T2D) and cardiovascular diseases (CVD) have always been major causes of death in Taiwan as well as many other countries in the world. They cause huge physical and economic burden not only to the patients themselves but also to the society. Therefore, the early detection and prevention are becoming important issues for health providers.

In the same time, the cluster of the CVD risk factors, including dyslipidemia, obesity, elevated blood pressure and hyperglycemia have been recognized as metabolic syndrome (MetS) and found to be highly correlated to the development of the aforementioned two diseases, each of the MetS components is also known to be an independent risk factor. However, there is no conclusive evidence to support which component outweighs other ones. This is not surprising since the importance is not expected to be the same in different age, sex and ethnicities.

It is well-known that the complications of T2D, namely macro- and microvascular diseases, are highly related to the level of plasma glucose concentration. This cause-effect relationship could be extrapolated to subjects with pre-diabetes. Interestingly, Countho et al. further showed that the increased incidence of cardiovascular events could start even when the fasting plasma glucose (FPG) is between 75 and 100 mg/dl among healthy middle-aged males. After this pilot study, three more consecutive studies confirmed this untoward effects of high-normal FPG. However, there was still inconsistency in these observations. For example, in the study von Gunten et al., a J-shape curve was noted rather than a straight line. To solve the controversy, larger cohorts and longitudinal studies are needed. Instead, previous studies have shown that, even within normal limits, higher FPG levels carry a higher risk of T2D development. However, their studies only involved middle-aged subjects, not focusing on elderly population.

Due to the National Health Insurance policy, the life expectancy of Taiwanese is much longer than before. Thus, the geriatric health problem becomes a challenge to the health providers. As aforementioned, both T2D and CVD are the leading causes of death, particularly in the elderly. Therefore, how to identify subjects under risks of these two diseases are even more crucial than in the younger population.

To our knowledge, there has been no longitudinal and large-cohort study focusing on the role of high-normal FPG on the future occurrence of T2D/CVD in the elderly population. In this study, we enrolled 18 287 subjects older than 60 years of age without any medications for hypertension, hyperglycemia and hyperlipidemia. Our purposes were first to test whether the untoward effects from the blood glucose could still be noted even in subjects with normal FPG. In the same time, a cut-off point was also identified which could be used for predicting higher chance to have either T2D or CVD in this cohort.

Materials and Methods
Subjects and sample collection
We enrolled subjects aged above 60 years (included) who underwent routine health checkups at the MJ Health Screening Center in Taiwan. MJ Health Screening Centers are privately owned chain of clinics located throughout Taiwan that provide regular health examinations to their members. All study participants were anonymous, and informed consents were obtained from all participants. Data were provided by MJ Health Screening Center for research purposes only, and the institutional review board of MJ Health Screening Center approved the study protocol. We randomly selected 57 517 records from MJ Health Screening Center’s database between 1999 and 2007. Subjects with past history of hypertension, T2D, cardiovascular events and taking medications for these diseases were all excluded (n = 16 479). In addition, we excluded the subjects with missing data of MetS components (n = 658) and FPG over 100 mg/dl (n = 22 113). Finally, a total of 18 287 subjects were eligible for analysis (Figure 1).

Analytic methods
Anthropometric measurements and general data
Senior nursing staff obtained subjects’ medical history, including information on any current medications, thorough questionnaire and complete physical examinations were performed. Waist circumference (WC) was measured horizontally at the level of the natural waist, which was identified as the level at the hollow molding of the trunk when the trunk was laterally concave. Body mass index was calculated as the subject’s body weight (kg) divided by the square of the subject’s height (m). Both systolic blood pressure (SBP) and diastolic blood pressure (DBP) were measured using nurse staff using standard mercury sphygmomanometers on the right arm of each subject when seated. After the subject had fasted for 10 h, blood samples were drawn from the antecubital vein for biochemical analysis. Plasma was separated from blood within 1 h and stored at −30°C until analysis for FPG and lipid profiles. FPG was measured using a glucose oxidase method (YSI 203 glucose analyzer, Yellow Springs Instrumentalists, Yellow Springs, USA). Total cholesterol and triglycerides (TG) were measured using a dry, multilayer analytical slide method with the Fuji Dri-Chem 3000 analyzer (Fuji Photo Film, Tokyo, Japan). Serum high-density lipoprotein cholesterol (HDL-C) and low-density lipoprotein cholesterol (LDL-C) concentration were analysed using an enzymatic cholesterol assay following dextran sulfate precipitation.

Definition of MetS, T2D and CVD
In this study, criteria for MetS were based on the modified Third Report of the Expert Panel on Detection, Evaluation, and Treatment of High Blood Cholesterol in Adults. Subjects with ≥3 of the following five criteria were diagnosed to have MetS: (i) WC ≥ 90 cm (Taiwanese males) and ≥ 80 cm (Taiwanese females), (ii) SBP ≥ 130 mmHg or DBP ≥ 85 mmHg, (iii) FPG ≥ 100 mg/dl, (iv) TG ≥ 150 mg/dl and (v) HDL-C ≤ 40 mg/dl (males) and ≤ 50 mg/dl (females). At the end of the follow-up, the occurrence of T2D was diagnosed based either on the FPG level (≥126 mg/dl), history of diabetes in the questionnaire or using oral anti-diabetic medication. In the same time, CVD was defined as any one of the following events reported: transient ischemic attack, ischemic or hemorrhagic stroke, acute myocardial infarction, unstable angina or angina pectoris, or having undergone percutaneous coronary intervention.
All statistical analyses were performed using SPSS 18.0 software (SPSS Inc., Chicago, IL). Data are presented as mean ± standard deviation. All data were tested for normal distribution with Kolmogorov-Smirnov test and for homogeneity of variances with Levene’s test. Data were log transformed before analysis if necessary.
data were not normally distributed. The t-test was used to evaluate the differences between the two groups. Correlations between MetS components and FPG were examined by Pearson correlation. Multivariate linear regression analysis was performed to further confirm if FPG was independently related to MetS after adjusting other confounding factors. There were two stages of the statistical evaluations. The first one was a cross-sectional stage (Stage 1). The purpose of this stage was to define the optimal FPG cut-off point, which was calculated by receiver operating characteristic (ROC) curve, to predict MetS in 8238 males and 10,049 females (MedCalc Software, Broekstraat, Mariakerke, Belgium). To validate these cut-off points in the second stage (longitudinal stage, Stage 2), subjects without MetS in the baseline were then grouped into low-FPG (baseline FPG < cut-off point) and high-FPG groups (baseline FPG > cut-off point). In Stage 2 of this study, 2337 males and 2702 females were followed up for an average of 3.8 years. Multivariable Cox regression analysis was performed to calculate the hazard ratio for developing T2D and CVD between these two groups. Finally, the Kaplan–Meier plot and log rank test were used to examine the differences of future T2D and CVD percentage. The details of the grouping methods are shown in Figure 1. A P value (two-sided) <0.05 was considered to be significant. Potential confounding factors that might have affected FPG, such as age, sex, smoking, alcohol consumption and physical activity were adjusted before analysis.

**Results**

Table 1 shows demographic data of subjects with and without MetS during the baseline. Not surprisingly, all of the components were significantly different due to our grouping method except that there were no differences in plasma LDL-C in both gender and TC levels in females. Table 2 depicts the results of univariate and multivariate analysis between FPG and other MetS components. In general, WC, SBP, DBP, TG and LDL-C were all positively correlated with FPG in both genders in univariate analysis. HDL-C was negatively correlated with FPG only in females. After the confounding factors were adjusted in multivariate analysis, FPG still remained significantly correlated with WC, SBP and LDL-C in both genders and negatively correlated with HDL-C in females. Optimal cut-offs of FPG were determined by ROC curve analysis, the results of which are given in Table 3. It demonstrated an optimal FPG cut-off for MetS was 93 mg/dl and 92 mg/dl for males and females (P=0.014 and 0.001, respectively).

In the Stage 2, 305 males and 397 females developed CVD who were exercised originally without having MetS (26.89 and 29.21 per 1,000 person-years, respectively). In the same time, 116 and 177 females had T2D (10.24 and 12.61 per 1,000 persons-years, respectively). By using multiple Cox regression, the high FPG group had a higher chance of developing T2D after 3.8 years follow-up for both males and females. However, the similar higher probability of having CVD was only noted in females after adjusting other confounding factors (Table 4). With the similar method, other MetS components were also evaluated. In this way, we could compare the effect of each MetS component on the incidence of the two endpoints. Our results show that higher WC and TG levels, but not low HDL-C, were independent factors for development of T2D in both genders. Other than this, higher BP carried higher risk for T2D incidence only in females. For the CVD outcome, all abnormal MetS components carried higher risk for further development of CVD in females, but only higher WC and BP were significantly associated with CVD in males. Figure 2 shows the results of Kaplan–Meier analysis. Not surprisingly, both genders with low-FPG had a significantly lower incidence of T2D than

**Table 1. Demographic data for subjects with and without MetS**

<table>
<thead>
<tr>
<th>MetS (−)</th>
<th>MetS (+)</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Male</td>
<td></td>
<td></td>
</tr>
<tr>
<td>N</td>
<td>7166</td>
<td>1072</td>
</tr>
<tr>
<td>Age (years)</td>
<td>66.2 ± 5.8</td>
<td>66.4 ± 5.7</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td>22.2 ± 5.41</td>
<td>26.2 ± 2.76</td>
</tr>
<tr>
<td>WC (cm)</td>
<td>81.9 ± 8.3</td>
<td>92.5 ± 7.3</td>
</tr>
<tr>
<td>SBP (mmHg)</td>
<td>128.5 ± 19.8</td>
<td>142.5 ± 16.7</td>
</tr>
<tr>
<td>DBP (mmHg)</td>
<td>75.2 ± 11.3</td>
<td>82.1 ± 10.7</td>
</tr>
<tr>
<td>Fasting Plasma Glucose</td>
<td>92.8 ± 5.2</td>
<td>93.1 ± 5.4</td>
</tr>
<tr>
<td>Glucose (mg/dl)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total cholesterol (mg/dl)</td>
<td>197.8 ± 36.6</td>
<td>201.4 ± 38.3</td>
</tr>
<tr>
<td>HDL-C (mg/dl)</td>
<td>52.8 ± 14.2</td>
<td>37.5 ± 8.4</td>
</tr>
<tr>
<td>LDL-C (mg/dl)</td>
<td>123.7 ± 33.1</td>
<td>123.1 ± 34.9</td>
</tr>
<tr>
<td>Log TG</td>
<td>1.98 ± 0.19</td>
<td>2.28 ± 0.16</td>
</tr>
</tbody>
</table>

**Table 2. Univariate and multivariate analysis of fasting plasma glucose and MetS components**

<table>
<thead>
<tr>
<th>Univariate</th>
<th>Multivariate</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>R</td>
</tr>
<tr>
<td>Male</td>
<td></td>
</tr>
<tr>
<td>Age (years)</td>
<td>−0.019</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td>0.033</td>
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<tr>
<td>WC (cm)</td>
<td>0.061</td>
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<tr>
<td>SBP (mmHg)</td>
<td>0.052</td>
</tr>
<tr>
<td>DBP (mmHg)</td>
<td>0.054</td>
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<tr>
<td>HDL-C (mg/dl)</td>
<td>−0.010</td>
</tr>
<tr>
<td>LDL-C (mg/dl)</td>
<td>0.059</td>
</tr>
<tr>
<td>Log TG</td>
<td>0.029</td>
</tr>
<tr>
<td>Female</td>
<td></td>
</tr>
<tr>
<td>Age (years)</td>
<td>−0.001</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td>−0.005</td>
</tr>
<tr>
<td>WC (cm)</td>
<td>0.048</td>
</tr>
<tr>
<td>SBP (mmHg)</td>
<td>0.053</td>
</tr>
<tr>
<td>DBP (mmHg)</td>
<td>0.052</td>
</tr>
<tr>
<td>HDL-C (mg/dl)</td>
<td>−0.034</td>
</tr>
<tr>
<td>LDL-C (mg/dl)</td>
<td>0.071</td>
</tr>
<tr>
<td>Log TG</td>
<td>0.012</td>
</tr>
</tbody>
</table>

BMI, body mass index; Log TG, log transformation of triglyceride. Data are shown as Mean ± SD.
Discussion

In this study, we confirmed our hypothesis that subjects with high normal FPG still carry higher chance to have T2D and CVD in the first stage. Moreover, we identified the cut-off points and validated these values in a 3.8-year follow-up in males and females separately. To our knowledge, this is the first and only study focusing on this topic in the elderly.

The relationship between FPG and CVD

The relationships between T2D and CVD have been well-established. One of the most solid evidences came from the report of Haffner et al. which showed that T2DM patients bear the same chance of having future myocardial infarction as subjects with a past history of myocardial infarction. Interestingly, the same relationships between plasma glucose levels and complications could still be extrapolated into not only subjects with IFG but also with normal FPG. However, in subjects with normal FPG, this relationship becomes inconsistent in various ethnic and age groups. For example, in an early Asian and Australian study, the positive associations between FPG and CVD could be observed, but only in subjects <60 years old. In the same time, instead of a straight line, a J-shape correlation with a nidar at 85–99 mg/dl, could be noted between FPG and incidence of CVD in a longitudinal adult Korean study. On the other hand, results supporting our finding could also be noted. For instance, Shaye et al. showed that the risks of CVD start to increase from the 85–90 mg/dl. In this longitudinal study, they also found that age and sex were the first and second important determinants. Similarly, data from a recent-published meta-analysis are also in line with our findings. From these observations, we could draw the conclusion that the rule of ‘the lower the better’ is generally applicable to plasma glucose level. However, the discrepancies of different studies might attribute to several factors, including different ethnic population, definition or CVD endpoints, age and gender.

The exact mechanism of high normal FPG increases CVD still remains unknown. The ‘ticking clock’ hypothesis proposed by Haffner et al. might help to explain this phenomenon. It considers that the plasma glucose level is a continuous variable and the risk of CVD increases progressively even before the development of T2D. This is not totally surprising since that increased FPG is positively and independently correlated with each MetS components separately. These untoward higher MetS risks could be translated into the higher incidence of CVD. From this hypothesis, it is reasonable to postulate that subjects with high normal FPG concentrations also have more severe subclinical atherosclerotic process than those with low-normal. Data from a Brazilian non-diabetic study supported this assumption and showed that the higher normal FPG was associated with the severity of coronary artery calcium. However, some other researchers don’t agree with this theory and suggested that FPG per se only serves as a surrogate marker of IR and its related metabolic traits abnormality. These risk factors are the true causatilites for the development of CVD. To verify this, studies with large cohort and prospective design are needed.

In this study, the relationship between FPG and CVD was only in females, but not males, is of interest. Nevertheless, our finding is not unique. Similar result was also supported by the study of Levitzky et al. They found that the risk for coronary artery disease in females started at a lower glucose level than males. In the same time, another study also demonstrated that non-diabetic females had significantly higher markers for endothelial dysfunction. To explain this difference, both Muller et al. and Mosca et al. suggested that sex hormones may play a role.

The relationships between FPG and T2D

The result of this study also showed that there is a significant prediction for future T2D even when the FPG is below 100 mg/dl. There are several putative mechanisms to explain the higher risk for T2D in subjects with high normal FPG levels, which included impaired first phase insulin secretion, increased hepatic glucose output due to liver insulin resistance and their counterparts (P = 0.014 and 0.049 in males and females, respectively, Figure 2a). For future CVD, only females with low-FPG had a significantly lower incidence (P = 0.035, Figure 2b). This ability of prediction did not reach statistical significance in males.
diminished non-insulin-dependent glucose clearance in these subjects.\textsuperscript{33} Our study was the only focused on this issue, other similar studies also suggested the cut-off values, however, in different age and ethnic groups.\textsuperscript{7,11–14,34} For example, in a 6-year longitudinal study, Tirosh et al.\textsuperscript{34} showed that incidence begins to increase when FPG levels were between 87 and 90 mg/dl in young males (mean age of 32 years old). Similarly, in another study done in middle-aged adults (mean age of 57.5 years), subjects with FPG between 90 and 94 mg/dl had greater risk of T2D compared with those with FPG $<\!85$ mg/dl.\textsuperscript{13} The rest of studies had similar cut-off FPG levels to predict further T2D development with various age population.\textsuperscript{7,11,12,14} It is interesting to note that even with a little variation; the range of different cutoff values is quite narrow. As aforementioned, this discrepancy might be due to the differences in age, ethnic groups, inclusion criteria and whether to take medications or not.

There are still limitations in our study. First, the participants were enrolled from the private health screening centers. Therefore, these subjects were in better economic status. The results may not be able to represent the general population. Extrapolating our data should be exercised with caution. Second, we only had FPG levels and did not have post-prandial glucose levels, which might also be highly related to T2D and CVD. There are evidences suggest that, compared with the post-prandial level, FPG has less contribution to the occurrence of future T2D or CVD.\textsuperscript{35} However, the measurement of post-prandial glucose would be a challenge as it is difficult to standardize the meal and time before the measurement. Furthermore, some subjects may have diabetes at enrollment because diabetic patients may exhibit normal FPG and not fulfill the criteria for MetS. It made the interpretation of our results with caution. Furthermore, psychological factors did not extensively evaluate in the present study, which might be confounding factors to

Figure 2. Kaplan-Meier estimates of (a) male T2D, (b) female T2D, (c) male CVD and (d) female CVD during follow-up based on fasting plasma glucose at baseline.
influence the CVD incidence. Finally, the diagnosis of CVD events was only confirmed from the questionnaire, and we did not have fatal CVD events, these may decrease the importance of our study.

In conclusion, our findings suggest that high-normal FPG (>93 mg/dl for male and 92 mg/dl for female, respectively) confers higher risks in subjects ≥60 year old. In the same time, they bear higher risks to have T2D in both genders and CVD in only females. Our results further strengthen the hypothesis of ‘the lower the better’ of plasma glucose in the elderly.

Conflict of interest: None declared.

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