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Clinical utility of 30-min plasma glucose for prediction of type 2 diabetes among people with prediabetes: *Ancillary analysis of the diabetes community lifestyle improvement program*

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ABSTRACT

Aims: To examine the clinical utility of 30-min plasma glucose (30-min-PG) measurement during an oral glucose tolerance (OGTT) in predicting type 2 diabetes (T2DM).

Research design and methods: Data from a 3-year, randomized, controlled, primary prevention trial among 548 Asian Indians with prediabetes were analyzed. Participants underwent OGTT with PG measurements at fasting, 30-min, and 2-h at baseline and annually until the end of the study. Multivariable Cox regression models were constructed to calculate the risk of developing diabetes based on 30-min-PG levels. Improvement in prediction performance gained by adding an elevated level of 30-min-PG over prediabetic categories was calculated using the area-under-curve (AUC), net-reclassification (NRI), and integrated discrimination improvement (IDI) statistics.

Results: At the end of follow-up, 30.4% of individuals had been diagnosed with T2DM by ADA criteria. Based on the maximally selected log-rank statistics, the optimal 30-min-PG cut point for predicting incident T2DM was >182 mg/dl. Multivariable-adjusted Cox regression models showed an independent association between elevated 30-min-PG (>182 mg/dl) and incident diabetes (hazard ratio (95% CI): 1.85 [1.32, 2.59]; D_{xy} = 0.353, c-statistic = 0.676). The addition of an elevated 30-min-PG (>182 mg/dl) model significantly improved the prediction of diabetes (Δdeviance: −15.4; ΔAUC: 0.11; $NRI_{continuous}$: 0.51; IDI: 0.08) compared with IFG model alone) in individuals with prediabetes.

Conclusion: In prediabetic individuals, baseline 30-min-PG independently predicted T2DM and significantly improved reclassification and discrimination. Therefore, 30-min-PG should be considered as part of the routine testing in addition to FPG and 2-h-PG for better risk stratification.

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1. Introduction

Accurate identification of individuals at heightened risk of developing type 2 diabetes (T2DM) is pivotal for the prevention of both T2DM and its associated complications. Impaired fasting glucose (IFG; ≥100–125 mg/dl [5.6–7.0 mmol/l]) as defined by American Diabetes Association (ADA) and impaired glucose tolerance (IGT; 2 h-PG ≥ 140–199 mg/dl [7.8–11.0 mmol/l]) [1], both of which can appear in isolation (i-IFG or i-IGT) or in combination (combined glucose intolerance [CGI]) during the oral glucose tolerance test (OGTT), are well-recognized markers of elevated risk for future T2DM. Prospective epidemiological studies have shown that ~40% of people with prediabetes progress to T2DM over a 5–7-year follow-up period [2]. Therefore, in addition to prediabetes, other markers may be required to identify those at the highest risk for developing T2DM accurately.

Prospective epidemiological studies have consistently demonstrated that intermediary glucose measures, or 1-h time points during a standard OGTT [3,4], and morphological characteristics of the glucose curve (e.g. glucose peak time and size, or the number of peaks) during OGTT [5,6], are associated with heightened risk of incident diabetes and cardiovascular and overall mortality. However, only a few studies [7,8] have examined the utility of 30-min plasma glucose (30-min-PG) on predicting incident diabetes. The deranged 30-min-PG response reflects inadequate first-phase insulin response and is the earliest detectable defect of pancreatic β-cell function in individuals destined to develop T2DM [9,10]. Therefore, assessing the intermediary glucose measurements at 30-min may have added benefit for identifying high-risk individuals. It is especially true for South Asians, as they exhibit long-term pancreatic β-cell compensation for chronic insulin resistance from childhood, resulting in an inability to produce further β-cell compensation in response to compounding insulin resistance in their later life [11,12]. However, the practical implications of using 30-min-PG for prediction of T2DM are less clear. Therefore, the aims of the present study were to assess the predictive role of 30-min-PG for progression to T2DM in individuals with prediabetes and examine the added predictive benefit of elevated 30-min-PG on top of IFG or IGT.

2. Methods

2.1. Study participants

Eligibility criteria, study methods, and primary results of the Diabetes Community Lifestyle Improvement Program (D-CLIP) have been reported in detail elsewhere [13,14]. Briefly, D-CLIP was a prospective, parallel-group, randomized controlled trial in adults with prediabetes in south-east India recruited between September 2009 and February 2012. The primary cohort consisted of 578 overweight/obese Asian Indians with prediabetes. They were randomized into two study groups: group-1 received standard care advice at baseline (n = 295); group-2 received a step-up diabetes prevention program (n = 283) which included six months of group-based, culturally-tailored, lifestyle education classes plus metformin for participants who remained at highest risk of converting to diabetes at four months or later. The intervention classes followed a structured educational curriculum with 16 weekly active period classes followed by eight weeks of maintenance classes. The curriculum was adapted from the U.S. Diabetes Prevention Program [15] and designed to reduce diabetes incidence through weight-loss (≥7% weight of baseline weight), 150 min or more of moderate activity weekly, and reducing intakes of fat (<30% of total energy) and total calories. Intervention group participants were prescribed a low dose of metformin (dose: 500 mg- twice daily) by the study physicians if individuals remained with or progressed to CGI or i-IFG + H bA1c > 5.7% at four months or later.

For this study, we included 548 participants with complete OGTT data. They were followed-up at 6-monthly intervals for the duration of the study (total of 36 months). At the annual follow-up (years 1, 2, and 3), a standard OGTT was carried out with blood sampling at three intervals (fasting, 30 min, and 120 min). During the interim visits (4, 6, 18, 30 months), a venous fasting plasma glucose (FPG) was taken to minimize the study participants' discomfort. The primary outcome was incident diabetes, based on the ADA guidelines: plasma glucose ≥126 mg/dl (≥7.0 mmol/l) in the fasting state or ≥200 mg/dl (≥11.1 mmol/l) 2-h after a 75-g oral glucose load [1]. The study showed that expert recommendations of adding low-dose of metformin in a stepwise manner to a structured lifestyle education was an effective method for delaying incident diabetes among overweight Asian Indians with prediabetes (intervention (25.7%) vs. standard-care advice (34.9%), with a relative risk reduction (RR) of 32% (95%CI: 7–50).

2.2. Physical and analytical determinations

At each visit, anthropometric, hemodynamic, and lifestyle measures were performed by trained personnel under standardized conditions following WHO recommendations [16]. The participants wore light clothes for all these measurements. Body weight was measured to the nearest 0.5 kg and height to the nearest 0.1 cm. Body mass index (BMI; kg/m^2) was computed. Waist circumference was measured midway between the lower rib and the iliac crest using a measuring tape to the nearest 0.1 cm [16].

The OGTT with venous blood sampling at three intervals (fasting, and at 30-min and 2-hours after 75 g anhydrous glucose) was done at baseline and annual visits. Plasma glucose (sodium fluoride as a preservative, hexokinase method, a coefficient of variation [CV] < 3% at 180 mg/dl and 360 mg/dl) was measured at each visit. Serum fasting triglycerides (GPO-PAP method, CV < 2.0%; detection range: 4.42–1000 mg/dl), total cholesterol (CHOD-PAP method, CV < 2.0%; detection range: 3.1–801 mg/dl), and HDL cholesterol (HDL plus-third generation; CV < 3.0%; detection range: 3.1–120 mg/dl) were measured annually with appropriate quality controls in an automated autoanalyzer. Plasma insu-

lin estimation was carried out on the Elecsys® platform (CV < 3%; detection range: 1.39–6945 pmol/L; ELICA; Roche Diagnostics, Germany).

2.3. Ethics

The Institutional Review Boards (IRB-00016503) of Emory University (Atlanta, USA) and the Madras Diabetes Research Foundation (Chennai, India) reviewed and approved the study procedures and materials. Written informed consent was obtained from each participant before screening, baseline testing, and randomization. The trial was registered on clinicaltrials.gov (NCT01283308; last updated: October 31, 2016).

2.4. Statistical analyses

Descriptive statistics were presented according to the diagnosis of diabetes. Because the primary objective of this *post hoc* analysis was to assess the predictive power of 30-min-PG on incident diabetes, we considered both control and intervention groups as a single cohort. Unadjusted and adjusted (age, sex, parental history of diabetes, and study allocation group) differences in baseline characteristics were estimated with quantile regression models. In addition, we also stratified the analyses based on 30-min-PG values in individuals with different prediabetes categories.

Multivariable Cox regression models were used to determine the hazard ratios for T2DM associated with baseline 30-min-PG levels (as a continuous variable). To examine the association between 30-min-PG and incident T2DM, we used natural cubic splines to allow for potential nonlinear relationships between biomarker level and incident T2DM. Follow-up time was estimated as the time from study entry until diabetes diagnosis or last investigation. Proportional hazards model then were adjusted for the following: Model-1 was adjusted for baseline age, sex, and allocation group; Model 2 was adjusted for the variables in Model-1 + parental history of diabetes and baseline body mass index (BMI); Model-3 was adjusted for baseline systolic blood pressure (SBP), HDL cholesterol, and triglycerides concentrations in addition to the variables in Model-2; and Model-4 was adjusted for the variables in Model 3 plus baseline levels of FPG and 2-h-PG. The proportional hazard assumption was tested for all predictors and covariates in a multivariable model, using the Schoenfeld residuals regressed against follow-up time; no violation of proportionality was observed. The "surv_cutpoint" command of the "survminer" [17], R package was used to split 30-min-PG concentrations into high- or low-level groups based on the maximally selected log-rank statistics (*maxstat*). Moreover, we set the "minprop" parameter of the "surv_cutpoint" function (referring to the minimal proportion of observations per group) to 30% to reduce the occurrence of too few individuals in a certain group.

In addition to that, the association of 30-min-PG categories (high vs. low) and the incidence of T2DM were generated using Cox proportional hazards models, while adjusting for the same potential confounders as above. Since the glycemic variables are highly correlated, we tested the collinearity for each of the covariates in the Cox models using the collinearity diagnostics. We observed no evidence of multicollinearity between covariates for any of the fitted models (variance inflation factor <2 for all independent variables). Several risk factors affect the association between 30-min-PG levels and incident T2DM, particularly baseline BMI, intervention allocation, parental history of diabetes, and baseline prediabetic status (iIFG, iIGT, or CGI). Therefore, we performed a prespecified subgroup analysis to assess whether the relationship between 30-min-PG levels and the risk of incident diabetes was robust in the presence of potential confounders. Since new sample data is presently not available, we chose to internally validate the model using the validate function in Harrell's *rms* package in R [18]. To do this, we used an enhanced bootstrapping resampling method, which accounts for bias due to overfitting, or optimism, using the Efron method. The objective of this calibration method is to assess whether the 30-min-PG predicts diabetes in new samples as accurately as it did in our sample. The predictive performance of these models was evaluated using Harrell's c-statistic and Sommer's Dxy.

2.5. Enhanced prognostication of 30-min-PG in predicting diabetes

The ability of 30-min-PG to enhance prognostication in addition to IFG and IGT models was tested with the deviance analysis, the area under the receiver operating characteristic (ROC) curve, net reclassification index (NRI), and integrated discrimination improvement (IDI). For this analysis, the following models were tested: Model-1) traditional diabetes risk factor model containing IFG, age, sex, parental history of diabetes, HDL cholesterol, triglycerides, BMI, and SBP; Model-2) model-1 and IGT; Model-3) model-1 and 30-min-PG; and Model-4) model-2 + 30-min-PG. Enhancements in the predictive performance of models 2, 3, or 4 were compared with model 1 (IFG model) and tested using continuous NRI. The continuous NRI is advantageous as it does not depend on the choice of specific risk categories and any change in predicted risk in the correct direction is considered appropriate. The goodness-of-fit was assessed by the Hosmer-Lemeshow chi-square test. All estimates are reported with 95% CI. All analyses and visualizations were conducted in R version 3.5.1 (R foundation of statistical computing, Vienna, Austria) using the TableOne (version: 0.10.0) [19], survival (version: 2.37–7) [20], survminer (version: 0.3.1) [17], and predictABEL (version: 1.2–2) packages.

3. Results

3.1. Baseline characteristics of study population

The mean age was 44.6 years, with a moderate representation of females (n = 206, 37.5%) and a mean BMI of 27.9 kg/m^2 at baseline. Glucose levels at 30-min and FPG (r = 0.408) were moderately correlated, whereas there was a weaker association between 30-min-PG values and 2-hr-PG (r = 0.126). Table 1 depicts demographic, anthropometric, and metabolic characteristics for the entire cohort stratified by the presence or absence of incident diabetes after follow-up. Participants who progressed to T2DM had a higher prevalence of a paren-

Table 1 – Baseline demographics, anthropometric, and metabolic characteristics of participants based on diabetes status during follow-up.

n	Non-Progressors 381	Progressors 167
Gender (Female, n, %)	141 (37.0)	64 (38.3)
Positive family history of Diabetes (%)	211 (55.4)	104 (62.3)
Group (Intervention, n, (%))	199 (52.2)	69 (41.3)
Age (median [IQR])	44.28 [38.44, 51.02]	44.56 [37.82, 50.95]
BMI (median [IQR])	27.22 [25.01, 29.38]	27.83 [25.78, 30.60]
Adjusted*	Ref	0.76 [0.21, 1.43]
Waist Circumferance (median [IQR])	93.70 [88.20, 99.80]	95.40 [90.85, 100.40]
Adjusted*	Ref	1.90 [0.03, 3.00]
Systolic blood pressure (median [IQR])	123.00 [114.00, 133.00]	124.00 [116.00, 134.50]
Adjusted*	Ref	1.09 [-1.60, 4.56]
Diastolic Blood Pressure (median [IQR])	73.50 [68.50, 81.00]	76.00 [70.50, 81.50]
Adjusted*	Ref	2.69 [0.56, 4.43]
Fasting Plasma Glucose (median [IQR])	102.00 [96.00, 106.00]	106.00 [100.00, 113.00]
Adjusted*	Ref	4.83 [3.00, 6.02]
30-min Plasma glucose (median [IQR])	173.00 [157.00, 186.00]	182.00 [168.00, 196.50]
Adjusted*	Ref	8.65 [4.81, 15.14]
120-min Plasma glucose (median [IQR])	147.00 [131.00, 162.00]	158.00 [144.50, 175.50]
Adjusted*	Ref	11.78 [5.61, 15.43]
Fasting Insulin (median [IQR])	11.85 [8.57, 15.72]	13.00 [9.35, 18.25]
Adjusted*	Ref	1.05 [0.07, 2.28]
30-min insulin (median [IQR])	82.90 [55.80, 121.20]	65.05 [43.47, 90.68]
Adjusted*	Ref	−18.44 [-24.56, −10.74]
120-min insulin (median [IQR])	120.80 [82.30, 205.40]	122.50 [77.75, 184.75]
Adjusted*	Ref	−1.32 [-17.12, 11.60]
Total Cholesterol (median [IQR])	187.00 [163.00, 215.00]	184.00 [161.00, 201.50]
Adjusted*	Ref	−3.92 [-11.03, 4.04]
HDL-Cholesterol (median [IQR])	39.00 [35.00, 44.00]	37.00 [34.00, 42.50]
Adjusted*	Ref	−1.80 [-2.78, −0.27]
LDL-Cholesterol (median [IQR])	116.00 [100.00, 141.00]	113.00 [97.25, 130.75]
Adjusted*	Ref	−0.44 [-6.73, 5.43]
Triglycerides (median [IQR])	133.00 [100.00, 178.00]	136.00 [104.00, 177.00]
Adjusted*	Ref	8.33 [-3.53,18.46]

For the continuous variables, the median (Q1–Q3) is presented in the first row followed by the median (95% CI) adjusted (age, sex, and allocation group) differences in the second row. Categorical variables are expressed as n (%).

tal history of diabetes, higher BMI, waist circumference, diastolic blood pressure, fasting, 30-min, and 2-h PG, and increased fasting plasma insulin. Moreover, the levels of HDL-cholesterol and 30-min plasma insulin were lower in the T2DM group compared with those who did not progress to diabetes.

3.2. *Higher 30-min-PG levels are associated with increased type 2 diabetes incidence*

During a median follow-up time of 2.5 (IQR: 2.1–3.1) years, 167 (30.4%) individuals with prediabetes at baseline developed T2DM. Table 2 presents Cox proportional hazards models testing the effects of baseline 30-min-PG on the incidence of diabetes. Based on natural cubic splines analysis, there was no evident deviation from the linearity of baseline 30-min-PG concentrations in Cox's proportional hazards model (P = 0.27; supplemental Fig. 1). Overall, higher 30-min-PG levels at baseline were associated with an increase in incident diabetes (adjusted hazard ratio [aHR]: 1.21 per 1 SD increase [95%CI: 1.02–1.44]). Based on the maximally selected rank statistics function, the optimal cut point for 30-min-PG to predicts incident diabetes was 182 mg/dl (supplemental Fig. 2).

Subsequently, the cumulative event curves showed a higher risk of incident diabetes with higher 30-min-PG category (>182 mg/dl) compared with a lower threshold (≤182 mg/dl) and a consistent separation in risk among 30-min-PG groups over time (Fig. 1). The median time to develop diabetes was 4.2 years in the 30-min-PG ≤ 182 mg/dl group compared with 3.5 years in the 30-min-PG > 182 mg/dl group (P < 0.0001). The impact of 30-min-PG > 182 mg/dl group on the incidence of diabetes occurred early in the study (at 6 months), showing that its effect was rapid. In multivariable adjusted Cox models, participants in the highest 30-min-PG (>182 mg/dl) group were more likely to report a subsequent diagnosis of diabetes than those with 30-min-PG ≤ 182 mg/dl, with a HR of 1.85 (95% CI: 1.32, 2.59) after adjustment for baseline age, sex, parental history of diabetes, allocation group, BMI, HDL-cholesterol, TG levels, SBP, FPG, and 2-h PG levels (model 4; Table 2); the model was validated and corrected for over-optimism using k-fold cross-validation (k = 10) and bootstrap validation (D_{xy} = 0.353, c-statistic = 0.676).

Table 2 – Cox Proportional Hazard model for incident type 2 diabetes, stratified according to elevated 30-minutes plasma glucose levels.

	30-min-PGas a continuous variable	30-min-PG ≤ 182 mg/dl n = 355	30-min-PG > 182 mg/dl n = 193
Incidence of Type 2 Diabetes (n, %)	167 (30.4)	84 (23.7)	83 (43.0)
Model-1	1.42 [1.21, 1.67]	Ref	2.18 [1.60, 2.97]
Model-2	1.43 [1.22, 1.68]	Ref	2.19 [1.60, 2.98]
Model-3	1.42 [1.21, 1.66]	Ref	2.19 [1.60, 2.99]
Model-4	1.22 [1.03, 1.45]	Ref	1.84 [1.32, 2.58]

All the participants were prediabetes at baseline. Incidence of Type 2 Diabetes presented as number of events (percentage) during the follow-up. The association of 30-min-PG as a continuous variable (per-SD) and in categories (30-min-PG ≤ 182 mg/dl (reference category) vs. 30-min-PG > 182 mg/dl) with the incidence of diabetes were presented using Cox Proportional Hazard models. Model-1 was adjusted for baseline age, sex, and allocation group; Model 2 was adjusted for the variables in Model-1 + parental history of diabetes, and baseline body mass index; Model-3 was adjusted for the baseline systolic blood pressure, diastolic blood pressure, HDL cholesterol and triglycerides concentrations in addition to the variables in Model-2; Model-4 was adjusted for the variables in Model 3 plus baseline levels of fasting plasma glucose, 2-h postload glucose, and HbA1c levels.

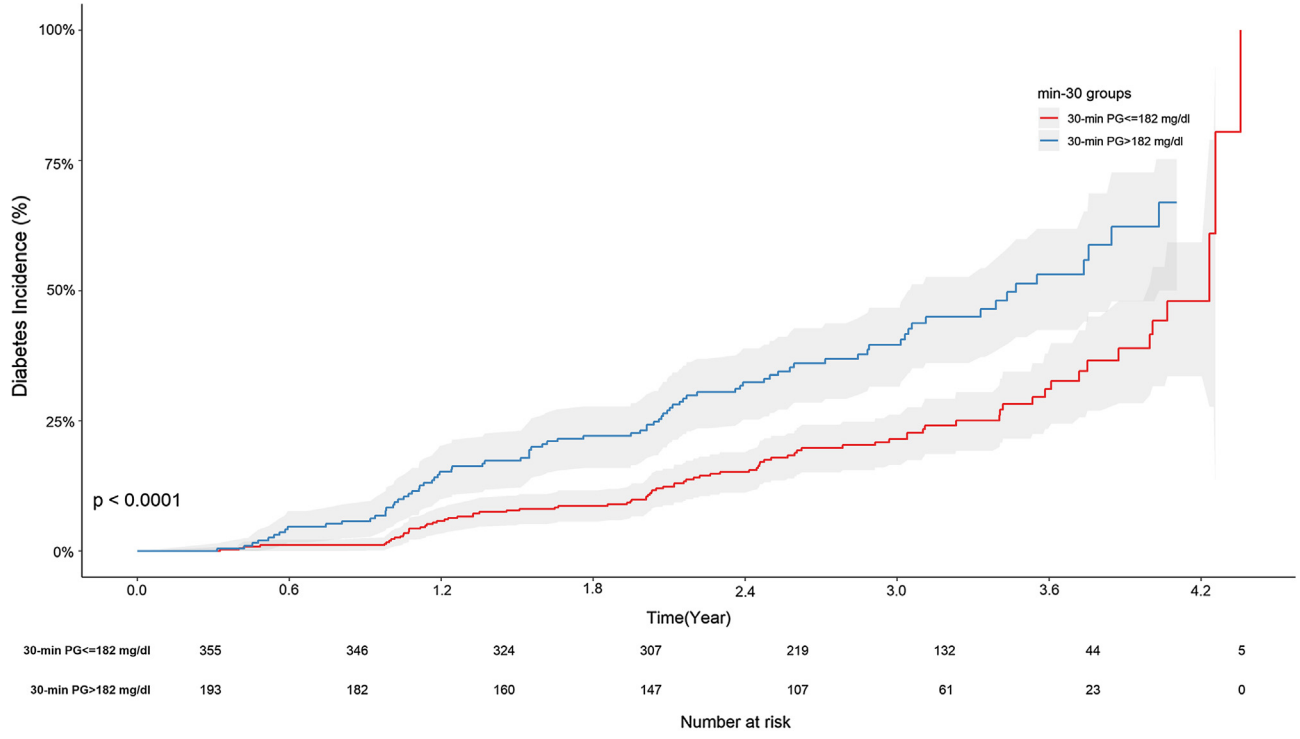

Fig. 1 – Cumulative incidence of diabetes by 30-min-Plasma Glucose from baseline to end of the study.

3.3. *Subgroup analysis*

We also examined whether a relationship between incident diabetes and baseline 30-min-PG existed among study subgroups (Fig. 2). We did not observe any differences in the demographic, anthropometric, and clinical characteristics between the two groups except FPG values. Associations of elevated 30-min-PG levels with incident diabetes were stronger among obese patients (BMI > 27.0 kg/m^2; aHR: 2.07 [95% CI: 1.37–3.12]), those with a parental history of diabetes (aHR: 2.30 [1.49–3.55]), in the standard care group (aHR: 1.89 [95%CI: 1.20–2.98]), and among those with combined glucose intolerance (aHR: 2.40 [95%CI: 1.56–3.70]) and iIGT (aHR: 2.36 [95%CI: 1.11–5.02]) at baseline. However, there were no significant interactions between 30-min-PG levels and baseline obesity status (P = 0.704), group allocation (P = 0.972), parental history of diabetes (P = 0.166) or prediabetes status at baseline (iIGT: P = 0.687; iIFG: P = 0.231 vs. CGI as ref).

3.4. *Predictive performance of 30-min-PG values in addition to FPG, and 2-h PG*

The model including 30-min-PG (deviance: 628) showed a significantly improved model fit compared with the traditional diabetes risk factor modeling including IFG (deviance: 646; Δdeviance: −17.7, P < 0.0001). The addition of 30-min-PG to the extended risk model including IFG and IGT (deviance: 620) significantly improved the model fit (deviance: 604; Δde-

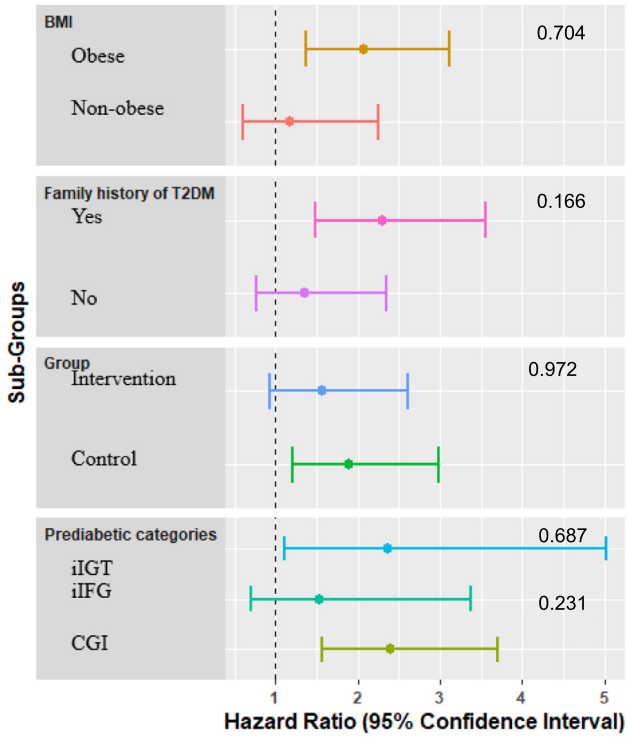

Fig. 2 – Subgroup analyses for effect modification by risk factors on risk for incident type 2 diabetes in participants with 30-min plasma glucose >182 mg/dl as compared to those with ≤182 mg/dl. Hazard ratio and 95% CI were obtained from multivariable Cox regression models adjusted for age, sex, BMI, systolic blood pressure, diastolic blood pressure, triglycerides, HDL, baseline prediabetic status. the variable was excluded from the model. CI, confidence interval.

viance: −15.9; P < 0.0001). Furthermore, the addition of 30-min-PG to the IFG and IGT model significantly improves the area under the ROC curve for the incidence of diabetes (Table 3; supplemental Fig. 3) and re-classification (NRI: 0.57 [0.39–0.74]; p-value: <0.0001; IDI: 0.08 [0.06–0.10]; p-value: <0.0001). The Hosmer-Lemeshow goodness-of fit test showed adequate calibration for all the models (all >P 0.05; supplemental Fig. 4).

4. Discussion

There are two significant findings in this analysis. Our study showed that 30-min-PG is a reliable and independent predictor of incident diabetes in individuals with prediabetes. Moreover, when 30-min-PG was added together with IFG or IGT, it improves the risk prediction of prediabetes progression to T2DM.

Although large scale epidemiological studies show an association between 1-h PG and diabetes [4], only a handful of studies show an association between 30-min-PG and incidence of diabetes [7,8,21]. Abdul-Ghani et al. [8] demonstrated that both elevated 30-min-PG and 1-h PG measurements were better predictors of future diabetes than fasting glucose in the San Antonio study. In a cross-sectional analysis in Chinese adults by Zhou et al., [21] the authors identified a 30-min-PG cut-off value of ≥175 mg/dl for prediabetes and ≥202 mg/dl cut-off for diabetes using ROC analysis. Additionally, a randomized controlled study among Asian Indians showed that 30-min-PG values in the upper tertile (>188 mg/dl) were independently associated with increased risk of developing diabetes compared with values in the lowest tertile (<163.0 mg/dl) [22] with an aHR of 1.44 (95% CI: 1.01–2.06) [7]. Our findings, however, contribute to new knowledge by identifying the optimal cut-point for 30-min-PG in predicting incident diabetes using robust statistical methodologies. Specifically, the results from our study suggest that elevated 30-min-PG of >182 mg/dl during an OGTT may identify a subgroup of individuals at increased risk for developing T2DM, likely due to decreased insulin sensitivity and pancreatic β-cell dysfunction. The sub-group analysis revealed that the association of 30-min-PG with incident diabetes was stronger among obese patients, in individuals with a positive family history of diabetes, in the standard care group, and in the presence of baseline combined glucose intolerance or iIGT. Importantly, the addition of 30-min-PG showed a significantly improved model fit and gain in classification accuracy for incident T2DM when added to an extended-adjustment model including prediabetes.

The term “prediabetes” is an umbrella term which encompasses a range of heterogeneous metabolic states with varying degrees of insulin resistance and pancreatic ß-cell dysfunction. Therefore, a better understanding of the etiology and pathophysiology of different prediabetic states might give a basis for the development of individualized prevention and treatment strategies for T2DM. Previously, using the retrospective samples from a real-world clinical setting, the STOP DIABETES study [22] showed the treatment benefit of 1-h-PG among individuals with NGT at baseline. As the peak glucose absorption occurs mostly at 30–60 min after ingesting a mixed meal, this period potentially represents an optimal period to detect the earliest evidence of metabolic dysfunction. Hence, glucose measurements at 30-min-PG provide an additional clinical advantage in further stratifying high-risk individuals even when applied as a filter within traditional glycaemic categories. Further investigations are warranted to validate our findings and explore the potential mechanisms.

To our knowledge, this is the first study to explore the clinical utility of 30-min-PG for predicting incident diabetes in South Asians with prediabetes. Our finding suggested a significant and valuable gain in model fit and classification accuracy when baseline 30-min-PG was added to a diabetes risk model including IFG and IGT. In clinical practice, prediabetes is not recognized as a clinical entity *per se* but rather as a risk factor for T2DM and cardiovascular disease progression [23]. Our study shows, for the first time, the ability of elevated 30-min-PG to separate high-risk from low-risk prediabetes categories. Although the potential contribution of 30-min-PG was previously appreciated, as shown by the inclusion of 30-min-PG in the 1979 National Diabetes Data Group criteria for classifying IGT, it was later deemed redundant in criteria set out by the WHO, which included 2 h-PG as the only post-challenge time point required for IGT classification. As only one-third of the individuals with the current prediabetic

Table 3 – Performance of adding 2-h PG and 30-min-PGconcentrations to the FPG PG for the prediction of incident diabetes.

	ROC			Net reclassification improvement – continuous		Integrated discrimination improvement	
	AUC	ΔAUC	P value	Mean (95%CI)	P value	Mean (95%CI)	P value
Model: 1 (IFG)	0.64 [0.59–0.69]	Ref		Ref	–	Ref	–
Model: 2 (IFG and IGT)	0.69 [0.64–0.74]	0.07	0.003	0.47 [0.30–0.64]	<0.0001	0.05 [0.03–0.07]	<0.0001
Model: 3 (IFG and 30-min-PG > 182 mg/dl)	0.68 [0.63–0.73]	0.04	0.089	0.42 [0.24–0.594]	<0.0001	0.03 [0.02–0.05]	<0.0001
Model: 4 (IFG + 30-min-PG + IGT)	0.72 [0.67–0.76]	0.11	0.0004	0.57 [0.39–0.74]	<0.0001	0.08 [0.06–0.10]	<0.0001

Model-1: includes baseline model (age, sex, parental history of diabetes, allocation group, body mass index, systolic blood pressure, diastolic blood pressure, triglycerides, and HDL-Cholesterol) with IFG.
Model-2: Baseline model + IFG + IGT.
Model-3: Baseline model + IFG + 30-min-PG > 182 mg/dl.
Model-4: Baseline model + FPG + 30-min-PG > 182 mg/dl + IGT.
Model performances were tested using logistic regression, where the "Model-1" was used as the reference.

criteria eventually convert to diabetes over 5–7 years, the additional glucose measurement at 30-minutes may proffer a basis for the further stratification of prediabetic individuals who are at extreme risk of developing diabetes.

This study has some limitations. At each timepoint, the OGTT was carried out once, which may have limited reproducibility, albeit, represents typical protocols in a real-world clinical setting. As the D-CLIP study only included overweight or obese South Asians, the findings of this study may not apply to those individuals with dysglycemia and healthy BMI. Although we controlled for the group allocation (control vs. intervention) in all the models, we did not additionally adjust for metformin, since most of the individuals allocated to the intervention were prescribed metformin (72%) by the end of follow-up. Furthermore, metformin should not alter the relationship between 30-min-PG and the incidence of diabetes. Also, we do not have information on 1-h PG in this cohort to make a direct comparison with 30-min-PG. However, previous work stemmed from Botnia studies [24,25] in Caucasians have shown that while 30-min-PG levels do not have as strong an association as 1 h PG based on ROC analysis in predicting diabetes, they do have a stronger association than the traditional glucose measures. Finally, although, we internally validated the clinical utility of 30-min-PG with incident diabetes using k-fold cross-validation and bootstrap to correct for over-optimism, our findings should be interpreted with caution as they are derived from Asian Indians with prediabetes without an external validation, which calls for further evaluations to assess the validity of 30-min-PG in other populations.

In summary, our data suggest that the 30-min-PG may represent an index of metabolic impairment, which is useful in clinical practice to identify individuals at high risk of developing T2DM. Predictive utility of glycemic thresholds at intermediary time points other than the traditional glycemic measures (fasting, 2-h plasma glucose) values should, therefore, be considered in clinical settings.

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Declaration of Competing Interest

The authors declared that there is no conflict of interest.

Data availability

The datasets generated during the current study are not publicly available but are available from the corresponding author on reasonable request.

Appendix A. Supplementary material

Supplementary data to this article can be found online at https://doi.org/10.1016/j.diabres.2020.108075.

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