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ADA HbA1c Diagnostic Criteria Fail to Identify Pre-diabetes and Diabetes in a Population of Chinese Adolescents and Young Adults at High Risk for Diabetes: a cross-sectional study

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Abstract

Objective We aimed to assess HbA1c for the diagnosis of pre-diabetes and diabetes in a population of Chinese youths at risk of metabolic syndrome.

Setting Beijing, China.

Participants A total of 581 subjects aged 14-28 year were recruited from the Beijing Child and Adolescent Metabolic Syndrome study. All participants underwent an oral glucose tolerance test (OGTT). Insulin sensitivity and β-cell function, and a number of cardiovascular disease risk factors were evaluated. Receiver operating characteristic curve (ROC) was performed to compare the screening efficacy.

Results Using OGTT data as a standard, the majority (70.0%, 7/10) of subjects with diabetes would have been diagnosed by HbA1c ≥ 6.5%. In contrast, only 28.1% (16/57) of subjects with pre-diabetes possessed elevated HbA1c's indicative of pre-diabetes, while the majority (68.4%) had normal HbA1c's. On the contrary, a total of 8.1% (39/479) of youths in the normal HbA1c category (<5.7%) and 21.3% in the pre-diabetes HbA1c category had pre-diabetes. For identifying prediabetes, the area under the curve (AUC) for HbA1c was 0.680 [95%CI 0.640-0.719]; the optimal threshold was 5.5%, with a sensitivity of 61.4% and specificity of 68.5%. For T2DM, the AUC for HbA1c was 0.970 [0.952-0.982], and the optimal threshold was 6.1% in, with sensitivity of 90.0% and a specificity of 98.7%. Compared with those HbA1c <5.5%, participants of HbA1c 5.5-6.1% showed lower disposition index and higher risk of being dyslipidemia (OR=1.61, [95% CI 1.10-2.37]) and metabolic syndrome (OR=2.09, [1.27-3.45]).

Conclusion The ADA's established HbA1c criteria for pre-diabetes and diabetes (5.7% and 6.5%) may not be appropriately applied to adolescents and young adults in the Chinese population. Our findings suggest that those with HbA1c of 5.5-6.1% already exhibit impaired β-cell function and increased cardio-metabolic risk factors, and may warrant intervention.

Key terms: HbA1c; Diabetes; Pre-diabetes; Metabolic syndrome; adolescents

Strengths and limitations of this study

This study included a well-characterized cohort of adolescents and young adults at risk for diabetes.

All these individuals have undergone an oral glucose tolerance test to evaluate their alterations in insulin sensitivity and β-cell function.

To our knowledge, this is the first study to assesse the ADA HbA1c cutpoints for predicting diabetes or pre-diabetes against the gold-standard OGTT in a population of Chinese adolescents and young adult.

Abbreviations

BMI: body mass index; WC: waist circumference; FBG: Fasting blood glucose; SBP: Systolic blood pressure; DBP: Diastolic blood pressure; TG: Triglycerides; TC: Total cholesterol; LDL-C: Low-density lipoprotein cholesterol; HDL□C: High□density lipoprotein cholesterol; CRP: C-reacting protein; OGTT: Oral glucose tolerance test; INS: Insulin; ISI: Insulin sensitivity index; IGI: Insulinogenic index; DIO: Oral disposition index; IFG: Impaired fasting glucose; IGT: Impaired glucose tolerance; HOMA-IR: The index of homeostasis model assessment of insulin resistance; MS: Metabolic syndrome; T2DM: Type 2 diabetes; ROC: Receiver operating characteristic.

Introduction

The incidence of obesity has increased dramatically in recent decades in Chinese youths and adolescents. The Global Burden of Disease Study showed that the prevalence of overweight and obesity has increased from 1980 to 2013 in children and adolescents in developing countries, from 8.4% to 13.4%[1]. With increasingly obesity, prevalence of diabetes has increased substantially. The World Health Organization (WHO) data demonstrates that in 2014, 347 million people worldwide have diabetes[2]. In china, the nationwide survey by Yang et al. in 2010 showed that the prevalence of diabetes and pre-diabetes had reached 9.7% and 15.5% in adults, respectively[3]. Both pre-diabetes and type 2 diabetes (T2DM) have been emerged as early complications of childhood obesity [4]and clustered with other cardiovascular risk factors[5]. There is increasing concern that obese youth is at risk for the long-term complications like diabetes and cardiovascular disease. Thus, it is important to identify the risk population predisposed to developing diabetes and target them for early intervention. It was suggested that impaired fasting glucose (IFG) and impaired glucose tolerance (IGT) assessment in youth was important for early prevention T2DM. However, due to expense, long duration of tests, conducting an oral glucose tolerance test (OGTT) is often cumbersome for patient care or population-based studies[6]. Fasting blood glucose (FBG) has been used as an inexpensive alternative to the OGTT, but FBG is also associated with challenges, like the requirement for an 8-h fast. In a study of adolescents reported failing to follow it for diabetic screening[7].

HbA1c is increasingly used by primary care providers as the screening test of choice due to its many advantages including convenience of sampling, a better index of chronic glycemia, low intra-individual variability, and assay standardization[8]. In 2010, the American Diabetes Association (ADA)[9] suggested that HbA1c values of 5.7-6.4% established a diagnosis of pre-diabetes while a value of ≥6.5% defined diabetes. These recommendations are based on data in adults showing the relationship betweenHbA1C with the subsequent development of diabetes and microvascular complications. However, the disagreement still

continue of what HbA1c level should be used to define pre-diabetes, and at least three different cut offs, 6.0%[10], 5.7%[9]and 5.5%[11]have been recommended. Furthermore, it is unclear whether these HbA1c thresholds should be applied to adolescents and young adults, due to the paucity of longitudinal data which associate these cut points in youth with ensuing adverse cardio-metabolic outcomes. Until these long-term outcomes become available, pre-diabetes and diabetes can be defined alternately by assessment of pathophysiologic abnormalities associated with hyperglycemia such as decliningβ-cell function and insulin sensitivity[12]. Currently, studies in the Chinese pediatric population are lacking. Therefore, the aim of this study was to assess HbA1c as an instrument to establish a diagnosis of pre-diabetes and diabetes in a population of Chinese adolescents and young adults at high risk of diabetes.

Materials and methods

Subjects

Subjects were recruited from the cohort of Beijing Children and Adolescents Metabolic Syndrome study (BCAMS). The BCAMS is longitudinal cohort study of cardiovascular risk factors since childhood. Details in baseline study have been described previously[13 14]. Briefly, in 2004 a population-based survey was conducted in Beijing area with a representative sample (n = 19,593, 50% boys) of schoolchildren (aged 6–18 years). Total 4500 subjects were identified as high risk at baseline due to having one of the following: overweight defined by body mass index (BMI), elevated totalcholesterol(TC) ≥5.2 mmol/L, elevated triglyceride (TG) ≥1.7mmol/L or elevated FBG ≥5.6 mmol/L based on finger capillary blood tests. Current follow-up study began in 2012 (8 years after baseline). Subjects were recruited consecutively through various modalities (phone, text and email) and underwent medical examination at a center in Beijing Chaoyang Hospital. Total 581 subjects who completed medical examination were included in this analysis. In baseline, those lost to follow-up were relatively younger and thinner than those followed-up, however,

there were no significant difference in gender, puberty status, blood pressures, fasting TG, low-density lipoprotein cholesterol (LDL-C) and high-density lipoprotein cholesterol (HDL-C)and FBG levels (P>0.05). Signed informed consent was obtained from all participants and/or their parents or guardians through all the study processes. The study was approved by the Ethics Committee at the Beijing Chaoyang Hospital.

Clinical measurements

Height, weight, waist circumference (WC) and percent body fat (FAT%)were measured by trained field workers. Participants removed bulky clothing and shoes prior to measurements. Height was measured to the nearest 0.1 cm using a portable stadiometer. WC was measured midway between the lowest rib and the top of the iliac crest. Weight and FAT mass was measured to the nearest 0.1kg using a TANITA Body Composition Analyzer (ModelTBF-300A). Measurements of right arm systolic and diastolic blood pressure (SBP and DBP) were performed 3 times 10 minutes apart and the mean values of the latter two measurements were recorded. BMI was calculated as weight divided by height squared.

Laboratory measurements

Venous blood samples were collected after an overnight (≥ 12h) fast. An OGTT test using 75g glucose load was performed on each subjects. FBG, 0.5h-BG and 2h-BG were measured by hexokinase method. The concentrations of TG, TC and LDL-C were assayed using a standard enzymatic method. HDL-C was assessed using precipitate with phosphotungstic acid-Mg method. Serum C-reacting protein (CRP) was measured by immunoturbidimetric assay. Insulin concentrations were measured by monoclonal antibody-based sandwich enzyme-linked immunosorbent assays which was developed in the Key Laboratory of Endocrinology, Peking Union Medical College Hospital. The intra- and inter -assay coefficient of variations (CVs) for insulin were < 5.4% and < 9.0%, respectively. Insulin assay had no cross-reactivity to proinsulin (< 0.05%).HbA1c were assayed using the TOSOH G7 automatic analysis system with high pressure liquid chromatography.

Insulin resistance was estimated by following index: (1) the index of homeostasis model assessment of insulin resistance (HOMA-IR) as (fasting insulin mU/L) × (FBG mmol/L)/22.5 [15];(2) Insulin sensitive index (Matsuda Index), ISI (Matsuda)= 10,000/ (FBG × fasting plasma insulin)×(mean plasma glucose ×mean plasma insulin) [16]. Pancreatic β-cell function was assessed by(1)homeostasis model assessment of β-cell function (HOMA-β) [17]; (2)insulinogenic index (IGI=ΔInsulin30/ΔGlucose30); (3) the ratio of the total area under the insulin curve to the total area under the glucose curve (total AUC Insulin/Glucose) and (4) the oral disposition index (DIO =IGI×ISI), which is the product of insulin sensitivity and insulin secretion, yields a better measure of beta cell function [18 19].

Definitions

Glucose tolerance status were defined according to the American Diabetes Association in 2010 [9], a subject was classified as having pre-diabetes including IFG: FBG ≥5.6mmol/l to 7.0mmol/l, IGT: 2h-BG ≥ 7.8mmol/l to 11.1mmol/l. T2DM was diagnosed in patients with FBG ≥7.0mmol/l or OGTT 2h-BG ≥ 11.1mmol/l. Metabolic syndrome (MS) was diagnosed according to 2009 proposed harmonized criteria, if the subject had at least three of the following five components [20]: (1) central obesity: WC ≥ 90th percentile for age and sex in 10 -16 years, or ≥ 90 cm for male and ≥ 80 cm for female; (2) IFG, IGT or a diagnosis of diabetes; (3) elevated BP: ≥130/85 mmHg; (4) HDL-C < 1.03mmol/L in males, < 1.29 mmol/L in females and (5) TG ≥ 1.70mmol/L. According to Chinese age- and sex-specific BMI cutoffs[21], adolescents were classified as overweight if BMI was between the 85th and 95th percentile, and obese if BMI was above 95th percentile. Subjects older than 18 year-old were classified overweight if BMI ≥ 24 kg/m^2, or obese if BMI ≥ 28 kg/m^2.

Data analysis

All statistical analyses were carried out using the Statistical Package for Social Sciences (SPSS 19.0 for windows). Continuous variables were tested for normality using a Kolmogorov-Smirnov test. Non-normal

distribution values used in the analyses were log-transformed to improve normality. Results are expressed as mean ± standard deviation (SD). Group comparisons across three HbA1c categories were made with ANOVA with Bonferroni *post hoc* comparison test. Agreement between HbA1c, fasting glucose category and OGTT 2-h glucose was also assessed. K coefficients were reported. Receiver operating characteristic (ROC) curve analysis was performed for HbA1c and FBG to discriminate pre-diabetes from normal glucose tolerance (NGT) and T2DM, from NGT and IGT using a logistic procedure. Area under the ROC curve (AUC) was considered as an effective measure of inherent validity of a diagnostic test. The mean values of variables studied by analysis of variance. Multivariate logistic regression models were used to estimate ORs for IR, MS and its components. Level of significance was accepted as $P < 0.05$.

Results

Subjects characteristics

The mean age of the entire population was 20.2 ± 2.9 years (female 46.8%). The prevalence rates of obesity, high blood pressure, dyslipidemia and MS were 32.6%, 20.2%, 29.5% and 14.5%, respectively. Of 581 subjects, 18 refused to conduct 2h-OGTT. According to ADA recommendation of HbA1c criteria, the detection rates of T2DM and at high risk for diabetes were 1.5% (9/581) and 13.4% (78/581), whereas according to OGTT criteria, IFG 4.8% (28/581), IGT 6.2% (35/563), IFG and /or IGT 10.1% (57/563) and T2DM 1.7% (10/581).

Comparisons between HbA1c and fasting glucose

The average HbA1c level was 5.4 ± 0.6%. HbA1c was strongly positive correlated with FBG (r = 0.734, $P < 0.001$), 2h-BG (r = 0.694, $P < 0.001$), while modest negative correlated with ISI (r = -0.177, $P<0.001$), IGI (r = -0.258, $P < 0.001$) and DIO (r = -0.389, $P < 0.001$) (Table 1). There were also modest positive relationship between HbA1c and cardio-metabolic parameters like TG (r = 0.159, $P < 0.001$), TC (r = 0.157,

$P < 0.001$), LDL-C (r = 0.176, $P < 0.001$), HDL-C (r = -0.103 $P < 0.05$), SBP (r = 0.143, $P = 0.001$) and hsCRP (r = 0.111, $P < 0.05$). FBG showed the similar results, but no significant relationship was observed with hsCRP ($P = 0.125$).

The agreement among HbA1c, fasting glucose with OGTT were showed in Table 2. First, using OGTT data as a standard, the majority (7/10, 70.0%) of subjects with diabetes would have been diagnosed by HbA1c ≥ 6.5%. In contrast, only 25.7% (9/35) of subjects with IGT possessed elevated HbA1c's indicative of pre-diabetes, while the majority (68.6%) had normal HbA1c's. Second, the majority (87.6%) of the subjects with NGT were classified as HbA1c < 5.7%, but 12.4 % were classified with at risk for diabetes or T2DM. On the other hand, of those considered as having T2D by using 2h-OGTT, 3 of 10 (30.0 %) were missed by an HbA1c > 6.4%, whereas among those in pre-diabetes (IFG, IGT, IFG/IGT) category, 39 of 57 (68.4 %) were missed by HbA1c criteria.

However, regarding for fasting glucose, of those diagnosed with diabetes using OGTT, only 4 of 10 (40.0%) were identified by their FBG values, and among those with IGT, only 2 of 35 (5.7%) was identified as IFG. On the other hand, of those considered as having T2DM by using OGTT, 6 of 10 (60.0%) were missed by the FBG, whereas among those IGT, 33 of 35 (94.3%) were missed by the FBG. In the other words, compared with HbA1c, using FBG criteria would miss more of IGT and T2DM. k coefficients as shown in Table 2 also demonstrate a poor agreement between either HbA1c criteria (k = 0.21) or FBG (k = 0.16) with 2h-OGTT.

ROC curve analysis

The AUCs shown in Figure 1A and 1B represent the diagnostic accuracy of the HbA1c, compared with FBG, for IGT and T2DM, respectively. For IGT, the AUC for HbA1c was 0.624 [95%CI 0.582-0.664] and the AUC for FBG was 0.663[0.576-0.749]. The optimal threshold of HbA1c was 5.5%, with a sensitivity of 42.9% and specificity of 78.6%. In the T2DM category, the AUC for HbA1c was 0.970 [0.952-0.982], and

the AUC for IFG was 0.789 [0.706-0.872]. The optimal threshold of HbA1c was 6.1% in identifying T2DM, with a sensitivity of 90.0% and specificity of 98.7%.

Giving that the status of IFG and IGT was largely poor agreement, we classified pre-diabetes by combined IFG and IGT, and the HbA1c test performance was further evaluated by ROC. As shown in Figure 1C and Table 3, the AUC of HbA1c for pre-diabetes was 0.680 (95%CI 0.640-0.719), and the optimal threshold of HbA1c was still 5.5%, with a sensitivity of 61.4% and specificity of 68.5%. Moreover, as shown in figure 1and Table 3, if ADA HbA1c criteria were evaluated, the sensitivity of the point for diagnosis of pre-diabetes in ROC analysis was decreased almost one-half, while the specificity was increased modestly.

Comparisons of metabolic characteristics according to different HbA1c criteria

To compare the metabolic characteristics between our proposed threshold of HbA1c and ADA criteria, we stratified the population according to HbA1c categories (Table 4). Age distribution, BMI, WC, DBP, FBG and 2h-BG (all $P < 0.05$) was different among the three categories detected on both ADA and our optimal thresholds, while HDL-C was not. As expected, there were more subjects (32.9% vs. 13.4%) classified as at risk for diabetes category based on HbA1c 5.5-6.1% than on ADA criteria of HbA1c 5.7-6.4%. Similarly, 16 (2.8%) vs. 9 (1.5%) subjects were considered having T2DM, respectively.

To compare in detecting β-cell dysfunction and insulin resistance by different HbA1c criteria, fasting and OGTT-derived measures were showed in Table 4. Subjects in the HbA1c 5.5-6.1% showed no difference in insulin resistance indices (HOMA-IR and ISI) comparing with HbA1c < 5.5%, neither in HbA1c 5.7-6.4% comparing with < 5.7%. However, subjects with HbA1c 5.5-6.1% showed a significant decrease of IGI comparing with HbA1c < 5.5%, and so did the DIO, which represents a measure of the insulin secretion adjusted for the insulin sensitivity. Notably, these situations were not prominent among groups classified by ADA criteria, especially regarding of IGI.

To further compare in detecting cardiovascular risk factors, as shown in Table 4 and 5, regardless of HbA1c criteria, lipids measures (TC, TG and LDL-C) were significantly higher in T2DM category than in normal or at risk for diabetes category, whereas hsCRP showed higher only in at risk for diabetes groups ($P < 0.05$). Moreover, high HbA1c was associated with a higher prevalence of obesity, high BP, IR and dyslipidemia. Of those with HbA1c 5.5-6.1%, 18.8 % had MS, compared with 10.5 % of those with HbA1c < 5.5%. In addition, as shown in Table 5, according to our HbA1c thresholds, HbA1c 5.5-6.1% versus HbA1c < 5.5% showed odds ratio of 1.61, 2.19 and 2.09 respectively, for developing dyslipidemia, insulin resistance and MS, which were more significant than ADA HbA1c 5.7-6.4% versus HbA1c < 5.7%.

Discussion

This cross sectional study demonstrates that an HbA1c of 5.7% and 6.4% had low sensitivity for classifying pre-diabetes (31.6%) and T2DM (63.6%). Our results suggest that, threshold of HbA1c was 6.1% for identifying T2DM, with a sensitivity of 81.8%and specificity of 98.8%, and 5.5% for identifying pre-diabetes with a specificity of 61.4% and sensitivity of 68.5%. We observed that the use of an HbA1c of 5.5% would largely improve the sensitivity of pre-diabetes and T2DM. Our data are in agreement with those reports of pediatrics [22] have concluded that HbA1c of 5.7-6.4% is inferior to detect pre-diabetes and T2DM.

Several pediatric studies have assessed ADA HbA1c cut points for predicting diabetes or pre-diabetes against the gold-standard OGTT and concluded that HbA1c is a poor predictor of pre-diabetes and T2DM in youth. Lee et al.[23] investigated both adolescents and adults, to diagnosis pre-diabetes and diabetes comparing HbA1c with 2h-BG. They found that it had poor sensitivity of 75% as HbA1c of 6.5%. Therefore, Laura M[24]put forward that prospective studies of pre-diabetes and T2DM in the obese pediatric population are especially needed to determine the HbA1c cutoff points, as well as other diagnostic measures, that best predict diabetes-related comorbid conditions later in life. The pediatric research, Paulina et al. [25] suggested

that HbA1c was 5.8% for identifying T2DM in obese children and adolescents. Compared with our study, the cut-off point of HbA1c to diagnosis T2DM was lower. Still, we can see that the criteria ADA-recommended cannot best serve us to make the diagnosis of diabetes or pre-diabetes in adolescents and young adults, especially in our population of high risk.

FBG has been used as an inexpensive alternative to the OGTT, especially for population screening of MS. In our study, we compared HbA1C versus FBG to detect dysglycemia. In the subjects categorized as pre-diabetes on the OGTT, 31.6% of them showed laboratory values indicative of at risk category or DM by the HbA1c, while only 5 (6.2%) were categorized as being IFG on the basis of FBG. Of the 10 classified with DM by OGTT, 7 of subjects were classified as having DM by an HbA1c, but only 4 (40.0%) would be indicated as having DM on the basis of FBG. In other words, 60.0% were missed by the FBG. It was subsequently suggested that HbA1c identified higher risk for diabetes than FBG. Similarly, a recent study in obese youth demonstrated that the HbA1c was relatively insensitive for detecting diabetes compared with FBG [25].

The debate over which test-HbA1c, FBG or 2h-OGTT is the better test for define glycemic abnormalities in youth ultimately requires decades of prospective studies to determine which test is more predictive of the cardiovascular and microvascular consequences. Until these long-term outcomes become available, pre-diabetes and diabetes can be defined alternately by pathophysiologic abnormalities associated with diabetes such as declining insulin sensitivity and β-cell function. Thus, in this study we compared the ADA criteria with our proposed cutoff point to detect those alterations in insulin sensitivity and β-cell function based on OGTT. We demonstrated the HbA1c 5.5-6.1% has clearly decreased in β-cell function (IGI, HOMA-β) as well as the DIO, which represents the insulin secretion relative to insulin sensitivity, is an established metabolic predictor of progression to diabetes [26]. We found progressively declining DIO across the HbA1c from < 5.5% to 5.5-6.1% to > 6.1%. However, the ADA HbA1c criteria cannot detect the

difference in β-cell function (IGI) from HbA1c < 5.7% to 5.5-6.4%. This implies that our proposed threshold might be more rationale for defining diabetes risk.

Studies have shown childhood glucose abnormity was associated with increased prevalence of cardiovascular risk factors [5]. We also found that subjects of at risk for diabetes defined by our HbA1c threshold of defined had more common of dyslipemia and metabolic syndrome. In the HbA1c 5.5-6.1%, compared with those < 5.5%, elevated HbA1c was associated with known risk factors for cardiovascular disease, including waist circumference, DBP, TC, TG and LDL-C, hsCRP as well as a more than twofold increased risk of having IR. There is an evolving consensus that, HbA1c can identify a population with higher risk of microvascular and macrovascular complications.

Moreover, we studied the criteria for the diagnosis of pre-diabetes and T2DM in adolescents and young adults for the reason of worrying about complications which were increased rapidly. Adolescents with pre-diabetes or T2DM potentially face many years of hyperglycemia and cardiovascular disease, thus, may have an increased lifetime risk of developing complications. In fact, it was reported that a large proportion of American adolescents have microalbuminuria and cardiovascular risk factors at diagnosis of T2DM [27]. Thus, early screening and intervention may be particularly beneficial in this young population, although the evidence base for the cut points for high risk for diabetes in youth is even more arbitrary than in adults.

Our study also has methodological limitations should be acknowledged. There was relatively small size of individuals with diabetes by ADA criteria in the population of youth. Strengths include the well-characterized cohort of adolescents and young adults at risk for diabetes.

In conclusion, the ADA's established HbA1c criteria for pre-diabetes and diabetes (5.7% and 6.5%) may not be appropriately applied to adolescents and young adults, especially in the Chinese population. Our findings suggest that those with HbA1c of 5.5 - 6.1% already exhibit impaired β-cell function and increased cardio-metabolic risk factors, and may warrant intervention. In view of fact that the rationale of choice of

the cut point to define high risk must take into account of the cost in order to prevent diabetes, the association of these proposed dysglycemic thresholds with micro- and macro-vascular complications of diabetes requires further investigation.

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Contributors GL analyzed data and drafted the manuscript; LWH, YHW, JLF and YL contributed to data collection; YLZ contributed to the data analysis and revised the manuscript; SMW contributed to the data interpretation and reviewed/edited the manuscript. ML contributed to the concept, design of the study,analyzed the data and revised the manuscript. SG was responsible for the concept, design, and data collection in the BCAMS follow-up study, and contributed to acquisition and interpretation of the data, and revised the manuscript.

Sharing statement Additional details on data presented in the current study are available by emailing liming@pumch.cn.

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Competing interests None declared.

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Table 1. Correlation of HbAc1, glucose with cardiometabolic risk parameters

	HbA1c	**FBG**	**2h-BG**
HbA1c (%)	1	0.734**	0. 694**
FBG (mmol/l)	0.734**	1	718**
2h-BG (mmol/l)	0. 694**	0.718**	1
TG (mmol/l)	0.159**	0.182**	0.196**
LDL-C (mmol/l)	0.176**	0.108**	0.152**
TC (mmol/l)	0.157**	0.032	0.048
HDL-C (mmol/l)	-0.103*	-0.095*	-0.102*
SBP (mmHg)	0.143**	0.151**	0.219**
DBP (mmHg)	0.209**	0.63**	0.238**
MS score	0.270**	0.215**	0.326**
Ln CRP (mg/l)[#]	0.112*	0.069	0.126**
Ln ISI	-0. 177**	-0.226**	-0.304**
Ln IGI	-0.258**	-0.213**	-0.282**
Ln DIO	-0.389**	-0.386**	-0.528**
Ln FINS (mU/L)	0.169**	0.182**	0.198**
Ln 0.5h-INS (mU/L)	-0.096*	-0.121**	-0.083
Ln 2h-INS (mU/L)	0.038	-0.025	0.357**

Abbreviations: FBG: Fasting blood glucose; TG: Triglycerides; TC: Total cholesterol; LDL□C: Low□density lipoprotein cholesterol; HDL□C: High□density lipoprotein cholesterol; SBP: Systolic blood pressure; DBP: Diastolic blood pressure; CRP: C-reacting protein; INS: insulin; ISI: insulin sensitivity index; IGI: insulinogenic index; DIO: oral disposition index and MS metabolic syndrome.

MS score: numbers of MS components

* $P < 0.05$; ** $P < 0.01$.

Table 2. The frequency of subjects with prediabetes and T2DM meeting the diagnostic criteria (HbA1c, FBG and 2h-BG after 75 g-OGTT)

OGTT	HbA1c *			Total	FBG #			Total
	NGT (< 5.7%)	At risk for diabetes (5.7-6.4%)	T2DM (> 6.4%)		NGT (< 5.6 mmol/l)	IFG (5.6-7.0 mmol/l)	T2DM (> 7.0 mmol/l)	
NGT	454 (87.6)	64 (12.4)	0	518	496 (95.9)	21 (4.1)	0	518
IGT	24(68.6)	9 (25.7)	2 (5.7)	35	33 (94.3)	2 (5.7)	0	35
T2DM	1(10.0)	2(20.0)	7 (70.0)	10	2(20.0)	4 (40.0)	4 (40.0)	10
Total	479 (85.1)	75 (13.3)	9 (1.6)	563@	531(94.3)	27 (4.8)	5 (0.9)	563@

Abbreviations: FBG: Fasting blood glucose; NGT: Normal glucose tolerance; IFG: Impaired fasting glucose; IGT: Impaired glucose tolerance and T2DM: Type 2 diabetes.

Numbers in brackets are percentages of horizontal total

*kappa coefficient 0.21; # kappa coefficient 0.16; @ 18 of 581 subjects disagreed to undergo 2h-OGTT.

Table 3. Test performance characteristics of specific HbA1c thresholds for detecting prediabetes and according to OGTT

HbA1c Threshold	Prediabetes (IFG + IGT)			T2DM		
	sensitivity	1-specificity	Sensitivity + specificity	sensitivity	1-specificity	Sensitivity + specificity
2.7	1.000	1.000	1.000	1.000	1.000	1.000
3.8	1.000	0.998	1.002	1.000	0.998	1.002
4.0	1.000	0.996	1.004	1.000	0.996	1.004
4.1	1.000	0.994	1.006	1.000	0.995	1.005
4.3	1.000	0.992	1.008	1.000	0.993	1.007
4.5	1.000	0.990	1.010	1.000	0.991	1.009
4.7	0.982	0.986	0.997	1.000	0.986	1.014
4.8	0.982	0.972	1.011	1.000	0.973	1.027
4.9	0.982	0.950	1.033	1.000	0.953	1.047
5.0	0.965	0.911	1.054	1.000	0.917	1.083
5.1	0.895	0.853	1.042	1.000	0.857	1.143
5.2	0.877	0.764	1.113	1.000	0.776	1.224
5.3	0.825	0.615	1.210	1.000	0.635	1.365
5.4	0.702	0.452	1.250	1.000	0.476	1.524
5.5	0.614	0.315	1.300	1.000	0.344	1.656
5.6	0.491	0.200	1.292	0.900	0.228	1.672
5.7	0.316	0.115	1.201	0.900	0.136	1.764
5.8	0.175	0.058	1.117	0.900	0.071	1.829
5.9	0.123	0.026	1.097	0.900	0.036	1.864
6.0	0.070	0.014	1.056	0.900	0.020	1.880
6.1	0.070	0.006	1.064	0.900	0.013	1.887
6.2	0.070	0.004	1.066	0.800	0.011	1.789
6.3	0.053	0.004	1.049	0.800	0.009	1.791
6.4	0.035	0.002	1.033	0.800	0.005	1.795
6.5	0.035	0.000	1.035	0.700	0.004	1.696
6.8				0.700	0.000	1.700
7.1				0.600	0.000	1.600
7.4				0.500	0.000	1.500
8.4				0.400	0.000	1.400
9.5				0.300	0.000	1.300
10.4				0.200	0.000	1.200
12.5				0.100	0.000	1.100
14.8				0.000	0.000	1.000

Abbreviations: IFG: impaired fasting glucose; IGT: impaired glucose tolerance and T2DM: type 2 diabetes.

Table 4. Clinical features of the study population according to HbA1c categories

	ADA criteria			*P*	Our proposed criteria			*P*
	< 5.7%	**5.7-6.4%**	**≥ 6.5%**		**< 5.5%**	**5.5-6.1%**	**≥ 6.1%**	
N	494	78	9		374	191	16	
Age (years)	20.30 (0.13)	19.28 (0.33)†	22.11(0.86)§	0.001	20.38 (0.14)	19.67 (0.23)†	21.33 (0.90)	0.010
Sex (M/F)	253/241	49/29	6/3	0.115	189/185	108/83	11/5	0.176
BMI (kg/m^2)	25.41 (0.24)	27.53 (0.83)†	28.73 (2.82)	<0.001	25.03(0.26)	26.90(6.46)†	28.65(2.11)§	< 0.001
WC (cm)	84.4 (0.6)	89.7 (2.1)†	97.7 (7.8)§	<0.001	83.4 (0.7)	88.5 (1.12)†	94.0(5.6)§	< 0.001
SBP (mmHg) *	114.7(0.5)	114.5 (1.2)	127.5(3.6) §¶	0.002	114.6(0.6)	115.1 (0.8)	119.2 (2.7)	0.251
DBP (mmHg) *	72.9(0.39)	75.2(1.0)	81.7(2.9)§	0.003	72.3 (0.5)	74.9 (0.6) ‡	78.3 (2.2)‖	<0.001
FBG (mmol/l)*	4.85 (0.03)	4.99 (0.08)	9.24(0.23)§¶	< 0.001	4.82 (0.04)	4.94(0.06)	7.61 (0.19)‖&	< 0.001
2h-BG (mmol/l)*	5.87 (0.07)	6.46(0.18)†	16.43(0.51)§¶	< 0.001	5.80 (0.09)	6.15 (0.12)	12.89 (0.41)‖&	< 0.001
IFG, n (%)	16(3.2%)	8(10.3%)†	4 (44.4%)§¶	< 0.001	6 (1.6%)	15(7.9%))‡	7 (43.8%)‖&	< 0.001
IGT, n (%)	24(5.0%)	9 (12.0%)†	2 (22.2%)§¶	< 0.001	16(4.4%)	17 (9.2%))‡	2 (12.5%)‖&	< 0.001
Pre-diabetes, n (%)	39(8.1%)	16(21.3%)†	2(22.2%)§	< 0.001	22 (6.1%)	30(16.3%)‡	4 (25.0%)‖	< 0.001
T2DM, n (%)	1(0.2%)	2 (2.6%)	7(77.8%)§¶	< 0.001	0(0%)	1(0.5%)	9 (56.3%)‖&	< 0.001
Ln FINS (mIU/L)#	1.92(0.03)	1.97(0.07)	2.64(0.21)§¶	0.003	1.92(0.03)	1.95(0.04)	2.13(0.16)	0.421
Ln 2h-INS (mU/L)#	3.58(0.03)	3.76(0.09)	3.73(0.30)	0.164	3.60(0.04)	3.60(0.06)	3.76(0.21)	0.778
Ln HOMA-IR#	0.38(0.02)	0.46(0.07)	1.58(0.22)§¶	< 0.001	0.38(0.03)	0.42(0.05)	0.91(0.17)‖&	0.007
Ln HOMA-β#	4.66(0.03)	4.63(0.07)	4.30(0.22)	0.280	4.68(0.03)	4.64(0.04)	4.09(0.16)‖&	0.002

Ln ISI[#]	1.81(0.02)	1.72(0.06)	0.95(0.21)§¶	< 0.001	1.81(0.03)	1.79(0.04)	1.45(0.15)‖&	0.068
Ln IGI[#]	0.25(0.04)	0.12(0.09)	-1.20(0.3)§¶	< 0.001	0.30(0.04)	0.14(0.06)‡	-1.14(0.22)‖&	< 0.001
Ln DIO[#]	2.06(0.03)	1.84(0.09)†	-0.21(0.32)§¶	< 0.001	2.11(0.04)	1.92(0.06)‡	0.38(0.21)‖&	< 0.001
LDL-C (mmol/l)*	2.50(0.03)	2.62(0.08)	3.47(0.24)§¶	< 0.001	2.49(0.04)	2.58(0.05)	2.95(0.18)‖	0.022
HDL-C (mmol/l)*	1.43(0.01)	1.45 (0.03)	1.35 (0.10)	0.609	1.44(0.02)	1.42(0.02)	1.50(0.07)	0.576
TC (mmol/l)*	4.30(0.04)	4.56(0.10)†	5.55(0.33)§¶	< 0.001	4.30(0.05)	4.40(0.07)	5.12(0.24)‖&	0.003
TG (mmol/l)*	1.10(0.04)	1.29 (0.09)	2.24 (0.26)§¶	<0.001	1.08(0.04)	1.12(0.06)	2.30(0.20)‖&	< 0.001
Ln-CRP (mg/l)[#]	0.09(0.05)	0.45 (0.14)†	-0.03 (0.39)	0.043	0.015(0.061)	0.34(0.084)‡	0.283(0.302)	0.008
Obesity, n (%)	152(30.8%)	32 (41.0%)	5 (55.6%)	0.134	108(29.0%)	72(37.7%)	9(56.3%)	0.024
MS, n (%)	61(12.4%)	16(20.8%)	7 (77.8%)§¶	< 0.001	39(10.5%)	36(18.8%)‡	9(60.0%)‖&	< 0.001

Abbreviations: BMI: Body mass index; WC: Waist circumference; SBP: Systolic blood pressure; DBP: Diastolic blood pressure; FBG: Fasting blood glucose; IFG: impaired fasting glucose; IGT: impaired glucose tolerance; T2DM: type 2 diabetes; FINS: fasting insulin; HOMA-IR: the index of homeostasis model assessment of insulin resistance; ISI: insulin sensitivity index; IGI: insulinogenic index; DIO: oral disposition index; TG: Triglycerides; TC: Total cholesterol; LDL‐C: Low‐density lipoprotein cholesterol; HDL‐C: High‐density lipoprotein cholesterol; CRP: C-reacting protein and MS: metabolic syndrome.

*adjusted for age, sex and BMI; [#] Log transformed and adjust for age, sex and BMI; Data were shown as mean (SE) or number (percentage).

†: < 5.7% vs. 5.7-6.4%; §: < 5.7% vs. > 6.4%; ¶: 5.7%-6.4% vs. > 6.4%;

‡: < 5.5% vs. 5.5-6.1%; ‖: < 5.5% vs. > 6.1%; & : 5.5-6.1% vs. > 6.1%.

Table 5. Age and sex adjusted ORs and 95% CIs for IR, MS and its components according to HbA1c categories

	ADA Criteria			*p* for trend	Our proposed criteria			*P* for trend
	< 5.7%	**5.7-6.5%**	**≥ 6.5%**		**< 5.5%**	**5.5-6.1%**	**≥ 6.1%**	
Elevated BP	1(ref)	1.55 (0.88-2.74)	17.75** (3.26-96.76)	0.002	1(ref)	1.37 (0.89-2.15)	5.63** (1.88-16.65)	0.006
Dyslipidemia	1	1.53 (0.91-2.57)	5.02* (1.22-20.61)	0.026	1	1.61* (1.10-2.37)	3.34* (1.17-9.54)	0.008
IR	1	2.10** (1.25-3.55）	21.50** (2.56-180.56)	< 0.001	1	2.19** (1.46-3.29)	8.69* (2.54-29.70)	<0.001
MS	1	1.95* (1.04-3.64)	20.80** (4.15-104.22)	< 0.001	1	2.09** (1.27-3.45)	11.63** (3.85-35.10)	< 0.001

Abbreviations: IR: insulin resistance, defined by HOMA-IR > 2.6 and

MS: metabolic syndrome.

* vs ref. $P< 0.05$; ** vs ref. $P < 0.01$.

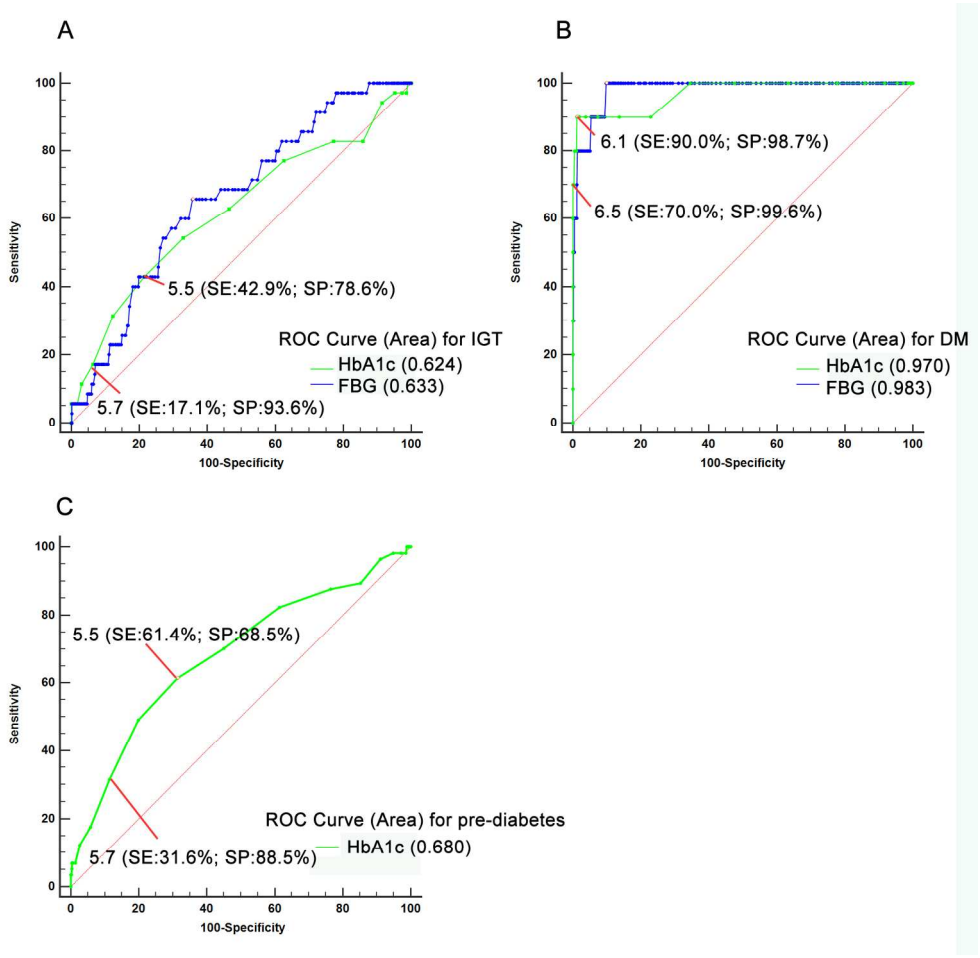

Figure 1. Comparison between the AUCs of the HbA1c and FBG for IGT (A) and T2DM (B), and the AUC of HbA1c for pre-diabetes (C). FBG means fasting blood glucose. SE means sensitivity. SP means specificity. The green discontinuous line indicates the curve defining the area for the HbA1c, and the blue continuous curve defines the area for FBG. Pre-diabetes was defined either by a FBG ≥5.6 mmol/l (IFG) or 2h-BG ≥7.8mmol/l (IGT).The red arrows indicate the different thresholds (sensitivity, specificity) of HbA1c.

173x164mm (300 x 300 DPI)

Section & Topic	No	Item	Reported on page #
TITLE OR ABSTRACT			
	1	Identification as a study of diagnostic accuracy using at least one measure of accuracy (such as sensitivity, specificity, predictive values, or AUC)	8
ABSTRACT			
	2	Structured summary of study design, methods, results, and conclusions (for specific guidance, see STARD for Abstracts)	2
INTRODUCTION			
	3	Scientific and clinical background, including the intended use and clinical role of the index test	4
	4	Study objectives and hypotheses	5
METHODS			
Study design	**5**	Whether data collection was planned before the index test and reference standard were performed (prospective study) or after (retrospective study)	5
Participants	**6**	Eligibility criteria	5
	7	On what basis potentially eligible participants were identified (such as symptoms, results from previous tests, inclusion in registry)	5
	8	Where and when potentially eligible participants were identified (setting, location and dates)	5
	9	Whether participants formed a consecutive, random or convenience series	5
Test methods	**10a**	Index test, in sufficient detail to allow replication	6-8
	10b	Reference standard, in sufficient detail to allow replication	6-8
	11	Rationale for choosing the reference standard (if alternatives exist)	6-8
	12a	Definition of and rationale for test positivity cut-offs or result categories of the index test, distinguishing pre-specified from exploratory	7-8
	12b	Definition of and rationale for test positivity cut-offs or result categories of the reference standard, distinguishing pre-specified from exploratory	7-8
	13a	Whether clinical information and reference standard results were available to the performers/readers of the index test	6-8
	13b	Whether clinical information and index test results were available to the assessors of the reference standard	6-8
Analysis	**14**	Methods for estimating or comparing measures of diagnostic accuracy	7-8
	15	How indeterminate index test or reference standard results were handled	7-8
	16	How missing data on the index test and reference standard were handled	5,7-8
	17	Any analyses of variability in diagnostic accuracy, distinguishing pre-specified from exploratory	7-8
	18	Intended sample size and how it was determined	5-6
RESULTS			
Participants	**19**	Flow of participants, using a diagram	5-6
	20	Baseline demographic and clinical characteristics of participants	8
	21a	Distribution of severity of disease in those with the target condition	8-9
	21b	Distribution of alternative diagnoses in those without the target condition	8-9
	22	Time interval and any clinical interventions between index test and reference standard	8-9
Test results	**23**	Cross tabulation of the index test results (or their distribution) by the results of the reference standard	8-10
	24	Estimates of diagnostic accuracy and their precision (such as 95% confidence intervals)	8-10
	25	Any adverse events from performing the index test or the reference standard	9-11
DISCUSSION			
	26	Study limitations, including sources of potential bias, statistical uncertainty, and generalisability	13
	27	Implications for practice, including the intended use and clinical role of the index test	13
OTHER INFORMATION			
	28	Registration number and name of registry	5
	29	Where the full study protocol can be accessed	5
	30	Sources of funding and other support; role of funders	14

STARD 2015

AIM

STARD stands for "Standards for Reporting Diagnostic accuracy studies". This list of items was developed to contribute to the completeness and transparency of reporting of diagnostic accuracy studies. Authors can use the list to write informative study reports. Editors and peer-reviewers can use it to evaluate whether the information has been included in manuscripts submitted for publication.

EXPLANATION

A **diagnostic accuracy study** evaluates the ability of one or more medical tests to correctly classify study participants as having a **target condition.** This can be a disease, a disease stage, response or benefit from therapy, or an event or condition in the future. A medical test can be an imaging procedure, a laboratory test, elements from history and physical examination, a combination of these, or any other method for collecting information about the current health status of a patient.

The test whose accuracy is evaluated is called **index test.** A study can evaluate the accuracy of one or more index tests. Evaluating the ability of a medical test to correctly classify patients is typically done by comparing the distribution of the index test results with those of the **reference standard**. The reference standard is the best available method for establishing the presence or absence of the target condition. An accuracy study can rely on one or more reference standards.

If test results are categorized as either positive or negative, the cross tabulation of the index test results against those of the reference standard can be used to estimate the **sensitivity** of the index test (the proportion of participants *with* the target condition who have a positive index test), and its **specificity** (the proportion *without* the target condition who have a negative index test). From this cross tabulation (sometimes referred to as the contingency or "2x2" table), several other accuracy statistics can be estimated, such as the positive and negative **predictive values** of the test. Confidence intervals around estimates of accuracy can then be calculated to quantify the statistical **precision** of the measurements.

If the index test results can take more than two values, categorization of test results as positive or negative requires a **test positivity cut-off**. When multiple such cut-offs can be defined, authors can report a receiver operating characteristic (ROC) curve which graphically represents the combination of sensitivity and specificity for each possible test positivity cut-off. The **area under the ROC curve** informs in a single numerical value about the overall diagnostic accuracy of the index test.

The **intended use** of a medical test can be diagnosis, screening, staging, monitoring, surveillance, prediction or prognosis. The **clinical role** of a test explains its position relative to existing tests in the clinical pathway. A replacement test, for example, replaces an existing test. A triage test is used before an existing test; an add-on test is used after an existing test.

Besides diagnostic accuracy, several other outcomes and statistics may be relevant in the evaluation of medical tests. Medical tests can also be used to classify patients for purposes other than diagnosis, such as staging or prognosis. The STARD list was not explicitly developed for these other outcomes, statistics, and study types, although most STARD items would still apply.

DEVELOPMENT

This STARD list was released in 2015. The 30 items were identified by an international expert group of methodologists, researchers, and editors. The guiding principle in the development of STARD was to select items that, when reported, would help readers to judge the potential for bias in the study, to appraise the applicability of the study findings and the validity of conclusions and recommendations. The list represents an update of the first version, which was published in 2003.

More information can be found on http://www.equator-network.org/reporting-guidelines/stard.

BMJ Open

Evaluation of ADA HbA1c Criteria in the Diagnosis of Pre-diabetes and Diabetes in a Population of Chinese Adolescents and Young Adults at High Risk for Diabetes: a Cross-sectional Study

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Evaluation of ADA HbA1c Criteria in the Diagnosis of Pre-diabetes and Diabetes in a Population of Chinese Adolescents and Young Adults at High Risk for Diabetes: a Cross-sectional Study

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Abstract

Objective We aimed to assess HbA1c for the diagnosis of pre-diabetes and diabetes in a population of Chinese youths at risk of metabolic syndrome.

Setting Beijing, China.

Participants A total of 581 subjects aged 14-28 years underwent evaluation including an oral glucose tolerance test (OGTT). Insulin sensitivity, β-cell function and a number of cardiovascular disease risk factors were evaluated. Receiver operating characteristic curves (ROC) were used to assess the screening efficacy of HbA1c.

Results Using OGTT data as a standard, the majority (70.0%, 7/10) of subjects with diabetes would have been diagnosed with HbA1c ≥ 6.5%. In contrast, only 28.1% (16/57) of subjects with pre-diabetes possessed elevated HbA1c's, while the majority (68.4%) had normal HbA1c's. On the contrary, a total of 8.1% (39/479) of youths in the normal HbA1c category (<5.7%) and 21.3% in the pre-diabetes category had pre-diabetes. In the ROC analysis, the area under the curve (AUC) for HbA1c identifying prediabetes, was 0.680 [95%CI 0.640-0.719]; the optimal threshold was 5.5%, with a sensitivity of 61.4% and specificity of 68.5%. For T2DM, the AUC for HbA1c was 0.970 [0.952-0.982], and the optimal threshold was 6.1%, with a sensitivity of 90.0% and a specificity of 98.7%. Applying these new cut-offs, pre-diabetic participants (HbA1c 5.5-6.1%) had lower disposition index and higher risk of dyslipidemia (OR=1.61, [95% CI 1.10-2.37]) and metabolic syndrome (OR=2.09, [1.27-3.45]) than those with normal HbA1c (<5.5%).

Conclusion The ADA's established HbA1c criteria for pre-diabetes and diabetes (5.7% and 6.5%) may not be appropriately applied to adolescents and young adults in China. Our findings suggest that those with HbA1c of 5.5-6.1% already exhibit impaired β-cell function and increased cardio-metabolic risk factors, which may warrant intervention.

Key terms: HbA1c; Diabetes; Pre-diabetes; Metabolic syndrome; adolescents

Strengths and limitations of this study

This study included a well-characterized cohort of adolescents and young adults at risk for diabetes.

All these individuals have undergone an oral glucose tolerance test to evaluate their alterations in insulin sensitivity and β-cell function.

This is the first study in a population of Chinese adolescents and young adults to assess the ADA's HbA1c cutpoints for predicting diabetes or pre-diabetes against the gold-standard OGTT.

However, there was relatively small size of individuals with diabetes by ADA criteria in the population of youth.

While the study cohort was a large population based sample, it may not be representative of the overall Chinese population as we chose to intensely study a subset at risk for the condition of interest (diabetes/pre-diabetes).

Abbreviations

BMI: body mass index; WC: waist circumference; FBG: Fasting blood glucose; SBP: Systolic blood pressure; DBP: Diastolic blood pressure; TG: Triglycerides; TC: Total cholesterol; LDL-C: Low-density lipoprotein cholesterol; HDL□C: High□density lipoprotein cholesterol; CRP: C-reacting protein; OGTT: Oral glucose tolerance test; INS: Insulin; ISI: Insulin sensitivity index; IGI: Insulinogenic index; DIO: Oral disposition index; IFG: Impaired fasting glucose; IGT: Impaired glucose tolerance; HOMA-IR: The index of homeostasis model assessment of insulin resistance; MS: Metabolic syndrome; T2DM: Type 2 diabetes; ROC: Receiver operating characteristic.

Introduction

The incidence of obesity has increased dramatically in recent decades among Chinese Chinese youths and adolescents. The Global Burden of Disease Study showed that the prevalence of overweight and obesity in children and adolescents in developing countries has increased from 8.4% in 1980 to 13.4% in 2013 [1]. World Health Organization (WHO) data that 347 million people worldwide have diabetes [2]. With the global surge in obesity, prevalence of diabetes has increased substantially. World Health Organization (WHO) data from 2014 estimated that 347 million people worldwide had diabetes [2]. A nationwide survey conducted by Yang et al. in 2010 showed that the prevalence of diabetes and pre-diabetes among adults in China had reached 9.7% and 15.5%, respectively [3]. As both pre-diabetes and type 2 diabetes (T2DM) have emerged as consequences of childhood obesity [4], the clustering of cardiovascular risk factors in this population heightens concern that obese children and young adults are at risk for complications of diabetes, specifically cardiovascular disease[5]. Thus, early identification of the population predisposed to developing diabetes is critically important if we are to target them for early intervention.

Screening for dysglycemia has traditionally focused on OGTT to identify diabetes, impaired fasting glucose (IFG) and impaired glucose tolerance (IGT). However, due to time, expense, and inconvenience, conducting an oral glucose tolerance test (OGTT) is often not feasible in patient care or population-based studies[6]. Fasting blood glucose (FBG) has been used as an inexpensive alternative to the OGTT, but FBG is also associated with challenges, like the requirement for an 8-h fast. In a study of diabetes screening practices among pediatric clinicians, a strong preference for non-fasting tests was evident [7].

HbA1c has become increasingly popular for diabetes screening among primary care providers due to its many practical advantages including: convenience of sampling, suitability as an index of chronic dysglycemia, low intra-individual variability, and propitious assay standardization [8]. In 2010, the American Diabetes Association (ADA)[9] suggested that HbA1c values of 5.7-6.4% established a diagnosis of

pre-diabetes while a value of ≥ 6.5% defined diabetes. These recommendations are based on data in adults showing the relationship between HbA1C and the subsequent development of diabetes and microvascular complications. However, it remains controversial what HbA1c level should be applied to the definition of pre-diabetes in children and adolescents, with at least three proposed thresholds: 6.0% [10], 5.7% [9]and 5.5% [11]. Furthermore, it is unclear at what ages these HbA1c thresholds should be applied, due to the paucity of longitudinal data in children (and even young adults) which associate these cut points with adverse cardio-metabolic outcomes. Until these long-term outcome data become available, pre-diabetes and diabetes can best be defined by their ability to identify pathophysiologic abnormalities associated with hyperglycemia such as decreased β-cell function and insulin sensitivity [12]. Currently, studies in the Chinese pediatric population are lacking. Therefore, the aim of this study was to assess HbA1c as an instrument to establish the diagnosis of pre-diabetes and diabetes in a population of Chinese adolescents and young adults at increased risk of diabetes.

Materials and methods

Subjects

Subjects were recruited from the cohort of Beijing Children and Adolescents Metabolic Syndrome study (BCAMS). The BCAMS is a longitudinal cohort study of cardiovascular risk factors since childhood. Details of the baseline study have been described previously [13-15]. Briefly, in 2004 a population-based survey was conducted in the Beijing area with a representative sample (n = 19,593, 50% boys) of schoolchildren (aged 6–18 years). Approximately 4500 subjects were identified as being at elevated risk for dysglycemia at baseline due to the presence of one of the following risk factors: overweight defined by body mass index (BMI), total cholesterol (TC) ≥5.2 mmol/L, triglyceride (TG) ≥1.7mmol/L or FBG ≥5.6 mmol/L based on finger capillary blood tests. A follow-up study began in 2012 (8 years after baseline), with Subjects recruited

consecutively through various modalities (phone, text and email) for medical examination at Beijing Chaoyang Hospital. A total of 581 subjects who completed medical examination are included in this analysis. Those lost to follow-up were relatively younger and thinner at baseline than those who did follow-up, however, there were no significant difference in gender, pubertal status, blood pressure, fasting TG, low-density lipoprotein cholesterol (LDL-C), high-density lipoprotein cholesterol (HDL-C) and FBG levels (P >0.05). The study was approved by the Ethics Committee at the Beijing Chaoyang Hospital and signed informed consent was obtained from all participants and/or their parents or guardians through all study phases. The BCAMS study was registered at www.clinicaltrials.gov (NCT03421444).

Patient and public involvement

No patients were involved in setting the research question or the outcome measures, nor were they involved in the design of this study. No patients were involved in the recruitment to and conduct of the study. There are no plans to disseminate the results of the research to study participants or the relevant patient community.

Clinical measurements

Height, weight, waist circumference (WC) and percent body fat (FAT%) were measured by trained field workers. Participants removed bulky clothing and shoes prior to measurements. Height was measured to the nearest 0.1 cm using a portable stadiometer. WC was measured midway between the lowest rib and the top of the iliac crest. Weight and FAT mass was measured to the nearest 0.1kg using a TANITA Body Composition Analyzer (ModelTBF-300A). Measurements of right arm systolic and diastolic blood pressure (SBP and DBP) were performed 3 times 10 minutes apart and the mean values of the latter two measurements were recorded. BMI was calculated as weight divided by height squared.

Laboratory measurements

Venous blood samples were collected after an overnight (≥ 12h) fast. An OGTT using 75g glucose load

was performed with plasma glucose levels in the fasting state (FBG), 0.5-hour (0.5hBG) and 2-hour (2hBG) measured using a hexokinase method. The concentrations of TG, TC and LDL-C were assayed using a standard enzymatic method. HDL-C was assessed with a phosphotungstic acid-Mg method. Serum C-reacting protein (CRP) was measured by immunoturbidimetric assay. Insulin concentrations were measured by monoclonal antibody-based sandwich enzyme-linked immunosorbent assays which was developed in the Key Laboratory of Endocrinology, Peking Union Medical College Hospital. The intra- and inter -assay coefficients of variation (CVs) for insulin were < 5.4% and < 9.0%, respectively, with no cross-reactivity to proinsulin (< 0.05%). HbA1c was assayed using the TOSOH G7 automatic analysis system with high pressure liquid chromatography. This assay is certified by the National Glycohemoglobin Standardization Program (NSPG).

Insulin resistance was estimated by following indeces: (1) the homeostasis model assessment of insulin resistance (HOMA-IR) [(fasting insulin mU/L) × (FBG mmol/L)/22.5]; (2) Insulin sensitive index (Matsuda Index), [ISI (Matsuda) = 10,000/ $\sqrt{((\text{FBG} \times \text{fasting plasma insulin}) \times (\text{mean plasma glucose} \times \text{mean plasma insulin}))}$] [16]. Pancreatic β-cell function was assessed by (1) homeostasis model assessment of β-cell function (HOMA-β) [(20 × fasting insulin)/(FPG-3.5)] [17]; (2) insulinogenic index (IGI=ΔInsulin30/ΔGlucose30); (3) the ratio of the total area under the insulin curve to the total area under the glucose curve (total AUC Insulin/Glucose) and (4) the oral disposition index (DI_O =IGI×ISI), which is the product of insulin sensitivity and insulin secretion [18 19].

Definitions

Dysglycemia (IFG, IGT, prediabetes, diabetes) was defined according to current American Diabetes Association guidelines [9]. Metabolic syndrome (MS) was diagnosed according to 2009 Joint Task Force harmonization criteria, with subjects exhibiting at least three of the following five components [20]: (1) central obesity: WC ≥ 90th percentile for age and sex in 10 -16 years, or ≥ 90 cm for male and ≥ 80 cm for female;

(2) IFG, IGT or diabetes; (3) BP: ≥130/85 mmHg; (4) HDL-C < 1.03mmol/L in males, < 1.29 mmol/L in females and (5) TG ≥ 1.70mmol/L. According to Chinese age- and sex-specific BMI cutoffs[21], adolescents were classified as overweight if BMI was between the 85th and 95th percentile, and obese if BMI was above 95th percentile. Subjects older than 18 year-old were classified overweight if BMI ≥ 24 kg/m^2, or obese if BMI ≥ 28 kg/m^2.

Data analysis

All statistical analyses were carried out using the Statistical Package for Social Sciences (SPSS 19.0 for windows). Continuous variables were tested for normality using a Kolmogorov-Smirnov test. Non-normal distribution values used in the analyses were log-transformed to improve normality. Results are expressed as mean ± standard deviation (SD). Group comparisons across three HbA1c categories were made with ANOVA with Bonferroni *post hoc* comparison test. Agreement between HbA1c, fasting glucose category and OGTT 2-h glucose was also assessed. K coefficients were reported. Receiver operating characteristic (ROC) curve analysis was performed for HbA1c and FBG to discriminate pre-diabetes from normal glucose tolerance (NGT) and T2DM, from NGT and IGT using a logistic procedure. Area under the ROC curve (AUC) was considered as an effective measure of inherent validity of a diagnostic test. The mean values of variables were studied by analysis of variance. Multivariate logistic regression models were used to estimate ORs for IR, MS and its components. Level of significance was accepted as $P < 0.05$.

Results

Subjects characteristics

The mean age of the entire population was 20.2 ± 2.9 years (female 46.8%). The prevalence rates of obesity, hypertension, dyslipidemia and MS were 32.6%, 20.2%, 29.5% and 14.5%, respectively. Of 581 subjects, 18 refused to conduct the 2h-OGTT. Using ADA criteria for HbA1c , the detection rates of diabetes

and pre-diabetes were 1.5% (9/581) and 13.4% (78/581), whereas according to OGTT criteria, IFG 4.8% (28/581), IGT 6.2% (35/563), IFG and /or IGT 10.1% (57/563) and T2DM 1.7% (10/581).

Comparisons between HbA1c and fasting glucose

The mean HbA1c level was 5.4 ± 0.6%. HbA1c showed a strong positive correlation to FBG (r = 0.734, $P < 0.001$) and 2h-BG (r = 0.694, $P < 0.001$), but a modest negative correlation with ISI (r = -0.177, $P<0.001$), IGI (r = -0.258, $P < 0.001$) and DI_O (r = -0.389, $P < 0.001$) (Table 1). There were also modest correlations between HbA1c and various cardio-metabolic parameters, such as TG (r = 0.159, $P < 0.001$), TC (r = 0.157, $P < 0.001$), LDL-C (r = 0.176, $P < 0.001$), HDL-C (r = -0.103 $P < 0.05$), SBP (r = 0.143, $P = 0.001$) and hsCRP (r = 0.111, $P < 0.05$). FBG showed similar correlation to these cardiometabolic parameters , except for hsCRP ($P = 0.125$).

The classification of subjects with regard to HbA1c and OGTT are shown in Table 2. First, using OGTT data as a standard, the majority (7/10, 70.0%) of subjects with diabetes would have been diagnosed by HbA1c ≥ 6.5%. In contrast, only 25.7% (9/35) of subjects with IGT possessed elevated HbA1c's indicative of pre-diabetes, while the majority (68.6%) had normal HbA1c's. Second, the majority (87.6%) of the subjects with NGT would be identified with HbA1c < 5.7%, while 12.4 % were classified with pre-diabetes or diabetes. On the other hand, of those considered to have diabetes by 2h-OGTT criteria, 3 of 10 (30.0 %) were missed by HbA1c, while those identified as pre-diabetic on an OGTT (i.e., IFG and/or IGT), 39 of 57 (68.4 %) were missed by HbA1c criteria.

However, of those diagnosed with diabetes using OGTT, only 4 of 10 (40.0%) were identified by their FBG values, and among those with IGT, only 2 of 35 (5.7%) were identified with IFG. So, using FPG criteria to identify dysglycemia would miss the majority of IGT and T2DM and using HbA1c was only moderately better (as demonstrated by the low k coefficients between either FBG (k=0.16) or HbA1c (k= 0.21) and OGTT.

ROC curve analysis

Figures 1A and 1B represent the diagnostic accuracy of the HbA1c and FBG, for IGT and diabetes identified by OGTT, respectively. The AUC for HbA1c was 0.624 [95% CI 0.582-0.664] and the AUC for FBG was 0.663 [0.576-0.749]. The optimal HbA1c threshold for identifying IGT was 5.5%, with a sensitivity of 42.9% and specificity of 78.6%. To identify diabetes, the AUC for HbA1c was 0.970 [0.952-0.982], and for IFG the AUC was 0.789 [0.706-0.872]. The optimal HbA1c threshold of 6.1%, identified diabetes with 90.0% sensitivity and 98.7% specificity.

In light of the inconsistency between IFG and IGT when identifying dysglycemia, we defined pre-diabetes as either IFG or IGT, and evaluated HbA1c test performance with ROC. As shown in Figure 1C and Table 3, the AUC of HbA1c for pre-diabetes was 0.680 (95%CI 0.640-0.719), and the optimal threshold of HbA1c was still 5.5%, with a sensitivity of 61.4% and specificity of 68.5%. Moreover, as shown in Figure 1and Table 3, compared with the ADA criteria for HbA1c (5.7%), lowering the HbA1c threshold to 5.5% doubles the sensitivity of this test, while only moderately affecting the specificity.

Comparisons of metabolic characteristics according to different HbA1c criteria

To compare the segregation of metabolic characteristics among groups identified by our proposed HbA1c thresholds versus the ADA criteria, we stratified the population according to HbA1c categories (Table 4). Age distribution, BMI, WC, DBP, FBG and 2h-BG (all $P < 0.05$) were all different among the three categories defined by either ADA or our proposed thresholds, while HDL-C was not. Not surprisingly, there were more subjects (32.9% vs. 13.4%) classified as pre-diabetic based on HbA1c 5.5-6.1% than by ADA criteria of 5.7-6.4%. Similarly, a greater number of subjects 16 (2.8%) vs. 9 (1.5%) would be considered to have T2DM by our criteria.

Table 4 compares the ability of these differing HbA1c strata to delineate levels of β-cell dysfunction and insulin resistance derived from FPG and OGTT measures. Subjects in the HbA1c 5.5-6.1% vs <5.5%

categories demonstrated no difference with regard to insulin resistance indices (HOMA-IR and ISI), and neither did those withHbA1c 5.7-6.4% comparing with those < 5.7%. However, subjects with HbA1c 5.5-6.1% showed a significantly lower IGI and DI_O compared to those with HbA1c < 5.5%. Notably, these differences were not as pronounced among groups classified using ADA criteria, especially with regard to IGI.

Regardless of the HbA1c thresholds employed, lipids measures (TC, TG and LDL-C) were significantly higher in diabetes than in normal or pre-diabetes categories, whereas hsCRP was highest in the pre-diabetic individuals ($P < 0.05$). Moreover, high HbA1c was associated with a higher prevalence of obesity, hypertension, IR and dyslipidemia. Of those with HbA1c 5.5-6.1%, 18.8 % had MS, compared with only 10.5 % of those with HbA1c < 5.5%. In addition, as shown in Table 5, applying our HbA1c thresholds, the odds ratios for dyslipidemia, insulin resistance and MS in pre-diabetic versus non-diabetic individuals were 1.61, 2.19 and 2.09 respectively, which is somewhat higher than if ADA criteria were employed.

Discussion

This cross sectional study demonstrates that an HbA1c of 5.7% and 6.4% had low sensitivity for classifying pre-diabetes (31.6%) and T2DM (63.6%). Our results suggest that, threshold of HbA1c was 6.1% for identifying T2DM, with a sensitivity of 81.8% and specificity of 98.8%, and 5.5% for identifying pre-diabetes with a specificity of 61.4% and sensitivity of 68.5%. We observed that the use of an HbA1c of 5.5% would largely improve the sensitivity of pre-diabetes and T2DM. Our data are in agreement with those reports of pediatrics [22] have concluded that HbA1c of 5.7-6.4% is inferior to detect pre-diabetes and T2DM.

Several pediatric studies have assessed ADA HbA1c cut points for predicting diabetes or pre-diabetes against the gold-standard OGTT and concluded that HbA1c is a poor predictor of pre-diabetes and T2DM in youth. Lee et al.[23] investigated both adolescents and adults, to diagnosis pre-diabetes and diabetes

comparing HbA1c with 2h-BG. They found that it had poor sensitivity of 75% as HbA1c of 6.5%. Therefore, Laura M [24] put forward that prospective studies of pre-diabetes and T2DM in the obese pediatric population are especially needed to determine the HbA1c cutoff points, as well as other diagnostic measures, that best predict diabetes-related comorbid conditions later in life. The pediatric research, Paulina et al. [25] suggested that HbA1c was 5.8% for identifying T2DM in obese children and adolescents. Compared with our study, the cut-off point of HbA1c to diagnosis T2DM was lower. Still, we can see that the criteria ADA-recommended cannot best serve us to make the diagnosis of diabetes or pre-diabetes in adolescents and young adults, especially in our population of high risk. These discrepancies between previous studies and ours might due to different age ranges, races and territory.

FBG has been used as an inexpensive alternative to the OGTT, especially for population screening of MS. In our study, we compared HbA1C versus FBG to detect dysglycemia. In the subjects categorized as pre-diabetes on the OGTT, 31.6% of them showed laboratory values indicative of at risk category or DM by the HbA1c, while only 5 (6.2%) were categorized as being IFG on the basis of FBG. Of the 10 classified with DM by OGTT, 7 of subjects were classified as having DM by an HbA1c, but only 4 (40.0%) would be indicated as having DM on the basis of FBG. In other words, 60.0% were missed by the FBG. It was subsequently suggested that HbA1c identified higher risk for diabetes than FBG. Similarly, Chan et al also showed that FPG performed poorly compared to HbA1c and OGTT and did not appear to have added value beyond HbA1c [26]. In contrast, a recent study in obese youth demonstrated that the HbA1c was relatively insensitive for detecting diabetes compared with FBG [25].

The debate over which test-HbA1c, FBG or 2h-OGTT is the better test for define glycemic abnormalities in youth ultimately requires decades of prospective studies to determine which test is more predictive of the cardiovascular and microvascular consequences. Until these long-term outcomes become available, pre-diabetes and diabetes can be defined alternately by pathophysiologic abnormalities associated with

diabetes such as declining insulin sensitivity and β-cell function. Thus, in this study we compared the ADA criteria with our proposed cutoff point to detect those alterations in insulin sensitivity and β-cell function based on OGTT. We demonstrated the HbA1c 5.5-6.1% has clearly decreased in β-cell function (IGI, HOMA-β) as well as the DIO, which represents the insulin secretion relative to insulin sensitivity, is an established metabolic predictor of progression to diabetes [27]. We found progressively declining DIO across the HbA1c from < 5.5% to 5.5-6.1% to > 6.1%. However, the ADA HbA1c criteria cannot detect the difference in β-cell function (IGI) from HbA1c < 5.7% to 5.5-6.4%. This implies that our proposed threshold might be more rationale for defining diabetes risk.

Studies have shown childhood glucose abnormity was associated with increased prevalence of cardiovascular risk factors [5]. We also found that subjects of at risk for diabetes defined by our HbA1c threshold of defined had more common of dyslipemia and metabolic syndrome. In the HbA1c 5.5-6.1%, compared with those < 5.5%, elevated HbA1c was associated with known risk factors for cardiovascular disease, including waist circumference, DBP, TC, TG and LDL-C, hsCRP as well as a more than twofold increased risk of having IR. There is an evolving consensus that, HbA1c can identify a population with higher risk of microvascular and macrovascular complications.

Moreover, we studied the criteria for the diagnosis of pre-diabetes and T2DM in adolescents and young adults for the reason of worrying about complications which were increased rapidly. Adolescents with pre-diabetes or T2DM potentially face many years of hyperglycemia and cardiovascular disease, thus, may have an increased lifetime risk of developing complications. In fact, it was reported that a large proportion of American adolescents have microalbuminuria and cardiovascular risk factors at diagnosis of T2DM [28]. Thus, early screening and intervention may be particularly beneficial in this young population, although the evidence base for the cut points for high risk for diabetes in youth is even more arbitrary than in adults.

Strengths include the well-characterized cohort of adolescents and young adults at risk for diabetes,

however, our study also has methodological limitations which should be acknowledged. Firstly, there was relatively small size of individuals with diabetes by ADA criteria in the population of youth, and further large studies are warranted to validate our findings. Secondly, we did not evaluate pubertal stage in this study, however, at the follow-up assessment 9 years later, the vast majority of the subjects would were over 16 years old, and at Tanner stage 5. Since puberty is associated with age, we have included age as a covariate when comparing the clinical features according to HbA1c categories, thus this may adjust the effect of pubertal stage to some degree. Thirdly, compared with our original population at baseline, the follow-up group is relatively small, which may introduce the potential for bias; however, there were no significant difference in gender, puberty status, and major cardiometabolic profiles at baseline between those followed-up and those lost to follow-up.

In conclusion, the ADA's established HbA1c criteria for pre-diabetes and diabetes (5.7% and 6.5%) may not be appropriately applied to adolescents and young adults, especially in the Chinese population. Our findings suggest that those with HbA1c of 5.5 - 6.1% already exhibit impaired β-cell function and increased cardio-metabolic risk factors, and may warrant intervention. Moreover, in view of fact that the rationale of choice of the cut point to define high risk must take into account of the cost in order to prevent diabetes, the association of these proposed dysglycemic thresholds with micro- and macro-vascular complications of diabetes requires further investigation.

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Contributors GL analyzed data and drafted the manuscript; LWH, YHW, JLF and YL contributed to data collection; YLZ contributed to the data analysis and revised the manuscript; SMW contributed to the data interpretation and reviewed/edited the manuscript. ML contributed to the concept, design of the study,

analyzed the data and revised the manuscript. SG was responsible for the concept, design, and data collection in the BCAMS follow-up study, and contributed to acquisition and interpretation of the data, and revised the manuscript.

Sharing statement Additional details on data presented in the current study are available by emailing liming@pumch.cn.

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Competing interests None declared.

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Table 1. Correlation of HbA1c, glucose with cardiometabolic risk parameters

	HbA1c	**FBG**	**2h-BG**
HbA1c (%)	1	0.734**	0. 694**
FBG (mmol/l)	0.734**	1	0.718**
2h-BG (mmol/l)	0. 694**	0.718**	1
TG (mmol/l)	0.159**	0.182**	0.196**
LDL-C (mmol/l)	0.176**	0.108**	0.152**
TC (mmol/l)	0.157**	0.032	0.048
HDL-C (mmol/l)	-0.103*	-0.095*	-0.102*
SBP (mmHg)	0.143**	0.151**	0.219**
DBP (mmHg)	0.209**	0.63**	0.238**
MS score	0.270**	0.215**	0.326**
Ln CRP (mg/l)[#]	0.112*	0.069	0.126**
Ln ISI	-0. 177**	-0.226**	-0.304**
Ln IGI	-0.258**	-0.213**	-0.282**
Ln DIO	-0.389**	-0.386**	-0.528**
Ln FINS (mU/L)	0.169**	0.182**	0.198**
Ln 0.5h-INS (mU/L)	-0.096*	-0.121**	-0.083
Ln 2h-INS (mU/L)	0.038	-0.025	0.357**

Abbreviations: FBG: Fasting blood glucose; TG: Triglycerides; TC: Total cholesterol; LDL‐C: Low‐density lipoprotein cholesterol; HDL‐C: High‐density lipoprotein cholesterol; SBP: Systolic blood pressure; DBP: Diastolic blood pressure; CRP: C-reacting protein; INS: insulin; ISI: insulin sensitivity index; IGI: insulinogenic index; DIO: oral disposition index and MS metabolic syndrome.

MS score: numbers of MS components

* $P < 0.05$; ** $P < 0.01$.

Table 2. The frequency of subjects with prediabetes and T2DM meeting the diagnostic criteria (HbA1c, FBG and 2h-BG after 75 g-OGTT)

OGTT	HbA1c *			Total	FBG #			Total
	NGT (< 5.7%)	Pre-diabetes (5.7-6.4%)	T2DM (> 6.4%)		NGT (< 5.6 mmol/l)	IFG (5.6-7.0 mmol/l)	T2DM (> 7.0 mmol/l)	
NGT	454 (87.6)	64 (12.4)	0	518	497 (95.9)	21 (4.1)	0	518
IGT	24(68.6)	9 (25.7)	2 (5.7)	35	33 (94.3)	2 (5.7)	0	35
T2DM	1(10.0)	2(20.0)	7 (70.0)	10	2 (20.0)	4 (40.0)	4 (40.0)	10
Total	479 (85.1)	75 (13.3)	9 (1.6)	563@	532 (94.5)	27 (4.8)	4 (0.7)	563@

Abbreviations: FBG: Fasting blood glucose; NGT: Normal glucose tolerance; IFG: Impaired fasting glucose; IGT: Impaired glucose tolerance and T2DM: Type 2 diabetes.

Numbers in brackets are percentages of horizontal total

*kappa coefficient 0.21; # kappa coefficient 0.16; @ 18 of 581 subjects disagreed to undergo 2h-OGTT.

Table 3. Test performance characteristics of specific HbA1c thresholds for detecting prediabetes and according to OGTT

HbA1c Threshold	Prediabetes (IFG + IGT)			T2DM		
	sensitivity	1-specificity	Sensitivity + specificity	sensitivity	1-specificity	Sensitivity + specificity
2.7	1.000	1.000	1.000	1.000	1.000	1.000
3.8	1.000	0.998	1.002	1.000	0.998	1.002
4.0	1.000	0.996	1.004	1.000	0.996	1.004
4.1	1.000	0.994	1.006	1.000	0.995	1.005
4.3	1.000	0.992	1.008	1.000	0.993	1.007
4.5	1.000	0.990	1.010	1.000	0.991	1.009
4.7	0.982	0.986	0.997	1.000	0.986	1.014
4.8	0.982	0.972	1.011	1.000	0.973	1.027
4.9	0.982	0.950	1.033	1.000	0.953	1.047
5.0	0.965	0.911	1.054	1.000	0.917	1.083
5.1	0.895	0.853	1.042	1.000	0.857	1.143
5.2	0.877	0.764	1.113	1.000	0.776	1.224
5.3	0.825	0.615	1.210	1.000	0.635	1.365
5.4	0.702	0.452	1.250	1.000	0.476	1.524
5.5	0.614	0.315	1.300	1.000	0.344	1.656
5.6	0.491	0.200	1.292	0.900	0.228	1.672
5.7	0.316	0.115	1.201	0.900	0.136	1.764
5.8	0.175	0.058	1.117	0.900	0.071	1.829
5.9	0.123	0.026	1.097	0.900	0.036	1.864
6.0	0.070	0.014	1.056	0.900	0.020	1.880
6.1	0.070	0.006	1.064	0.900	0.013	1.887
6.2	0.070	0.004	1.066	0.800	0.011	1.789
6.3	0.053	0.004	1.049	0.800	0.009	1.791
6.4	0.035	0.002	1.033	0.800	0.005	1.795
6.5	0.035	0.000	1.035	0.700	0.004	1.696
6.8				0.700	0.000	1.700
7.1				0.600	0.000	1.600
7.4				0.500	0.000	1.500
8.4				0.400	0.000	1.400
9.5				0.300	0.000	1.300
10.4				0.200	0.000	1.200
12.5				0.100	0.000	1.100
14.8				0.000	0.000	1.000

Abbreviations: IFG: impaired fasting glucose; IGT: impaired glucose tolerance and T2DM: type 2 diabetes.

Table 4. Clinical features of the study population according to HbA1c categories

	ADA criteria			*P*	Our proposed criteria			*P*
	< 5.7%	**5.7-6.4%**	**≥ 6.5%**		**< 5.5%**	**5.5-6.1%**	**≥ 6.1%**	
N	494	78	9		374	191	16	
Age (years)	20.30 (0.13)	19.28 (0.33)†	22.11(0.86)§	0.001	20.38 (0.14)	19.67 (0.23)†	21.33 (0.90)	0.010
Sex (M/F)	253/241	49/29	6/3	0.115	189/185	108/83	11/5	0.176
BMI (kg/m^2)	25.41 (0.24)	27.53 (0.83)†	28.73 (2.82)	<0.001	25.03(0.26)	26.90(6.46)†	28.65(2.11)§	< 0.001
WC (cm)	84.4 (0.6)	89.7 (2.1)†	97.7 (7.8)§	<0.001	83.4 (0.7)	88.5 (1.12)†	94.0(5.6)§	< 0.001
SBP (mmHg) *	114.7(0.5)	114.5 (1.2)	127.5(3.6) §¶	0.002	114.6(0.6)	115.1 (0.8)	119.2 (2.7)	0.251
DBP (mmHg) *	72.9(0.39)	75.2(1.0)	81.7(2.9)§	0.003	72.3 (0.5)	74.9 (0.6) ‡	78.3 (2.2)‖	<0.001
FBG (mmol/l)*	4.85 (0.03)	4.99 (0.08)	9.24(0.23)§¶	< 0.001	4.82 (0.04)	4.94(0.06)	7.61 (0.19)‖&	< 0.001
2h-BG (mmol/l)*	5.87 (0.07)	6.46(0.18)†	16.43(0.51)§¶	< 0.001	5.80 (0.09)	6.15 (0.12)	12.89 (0.41)‖&	< 0.001
IFG, n (%)	16(3.2%)	8(10.3%)†	4 (44.4%)§¶	< 0.001	6 (1.6%)	15(7.9%))‡	7 (43.8%)‖&	< 0.001
IGT, n (%)	24(5.0%)	9 (12.0%)†	2 (22.2%)§¶	< 0.001	16(4.4%)	17 (9.2%))‡	2 (12.5%)‖&	< 0.001
Pre-diabetes, n (%)	39(8.1%)	16(21.3%)†	2(22.2%)§	< 0.001	22 (6.1%)	30(16.3%)‡	4 (25.0%)‖	< 0.001
T2DM, n (%)	1(0.2%)	2 (2.6%)	7(77.8%)§¶	< 0.001	0(0%)	1(0.5%)	9 (56.3%)‖&	< 0.001
Ln FINS (mIU/L)#	1.92(0.03)	1.97(0.07)	2.64(0.21)§¶	0.003	1.92(0.03)	1.95(0.04)	2.13(0.16)	0.421
Ln 2h-INS (mU/L)#	3.58(0.03)	3.76(0.09)	3.73(0.30)	0.164	3.60(0.04)	3.60(0.06)	3.76(0.21)	0.778
Ln HOMA-IR#	0.38(0.02)	0.46(0.07)	1.58(0.22)§¶	< 0.001	0.38(0.03)	0.42(0.05)	0.91(0.17)‖&	0.007
Ln HOMA-β#	4.66(0.03)	4.63(0.07)	4.30(0.22)	0.280	4.68(0.03)	4.64(0.04)	4.09(0.16)‖&	0.002

Ln ISI[#]	1.81(0.02)	1.72(0.06)	0.95(0.21)§¶	< 0.001	1.81(0.03)	1.79(0.04)	1.45(0.15)‖&	0.068
Ln IGI[#]	0.25(0.04)	0.12(0.09)	-1.20(0.3)§¶	< 0.001	0.30(0.04)	0.14(0.06)‡	-1.14(0.22)‖&	< 0.001
Ln DIO[#]	2.06(0.03)	1.84(0.09)†	-0.21(0.32)§¶	< 0.001	2.11(0.04)	1.92(0.06)‡	0.38(0.21)‖&	< 0.001
LDL-C (mmol/l)*	2.50(0.03)	2.62(0.08)	3.47(0.24)§¶	< 0.001	2.49(0.04)	2.58(0.05)	2.95(0.18)‖	0.022
HDL-C (mmol/l)*	1.43(0.01)	1.45 (0.03)	1.35 (0.10)	0.609	1.44(0.02)	1.42(0.02)	1.50(0.07)	0.576
TC (mmol/l)*	4.30(0.04)	4.56(0.10)†	5.55(0.33)§¶	< 0.001	4.30(0.05)	4.40(0.07)	5.12(0.24)‖&	0.003
TG (mmol/l)*	1.10(0.04)	1.29 (0.09)	2.24 (0.26)§¶	<0.001	1.08(0.04)	1.12(0.06)	2.30(0.20)‖&	< 0.001
Ln-CRP (mg/l)[#]	0.09(0.05)	0.45 (0.14)†	-0.03 (0.39)	0.043	0.015(0.061)	0.34(0.084)‡	0.283(0.302)	0.008
Obesity, n (%)	152(30.8%)	32 (41.0%)	5 (55.6%)	0.134	108(29.0%)	72(37.7%)	9(56.3%)	0.024
MS, n (%)	61(12.4%)	16(20.8%)	7 (77.8%)§¶	< 0.001	39(10.5%)	36(18.8%)‡	9(60.0%)‖&	< 0.001

Abbreviations: BMI: Body mass index; WC: Waist circumference; SBP: Systolic blood pressure; DBP: Diastolic blood pressure; FBG: Fasting blood glucose; IFG: impaired fasting glucose; IGT: impaired glucose tolerance; T2DM: type 2 diabetes; FINS: fasting insulin; HOMA-IR: the index of homeostasis model assessment of insulin resistance; ISI: insulin sensitivity index; IGI: insulinogenic index; DIO: oral disposition index; TG: Triglycerides; TC: Total cholesterol; LDL‐C: Low‐density lipoprotein cholesterol; HDL‐C: High‐density lipoprotein cholesterol; CRP: C-reacting protein and MS: metabolic syndrome.

*adjusted for age, sex and BMI; [#] Log transformed and adjust for age, sex and BMI; Data were shown as mean (SE) or number (percentage).

†: < 5.7% vs. 5.7-6.4%; §: < 5.7% vs. > 6.4%; ¶: 5.7%-6.4% vs. > 6.4%;

‡: < 5.5% vs. 5.5-6.1%; ‖: < 5.5% vs. > 6.1%; & : 5.5-6.1% vs. > 6.1%.

Table 5. Age and sex adjusted ORs and 95% CIs for IR, MS and its components according to HbA1c categories

	ADA Criteria			*p* for trend	Our proposed criteria			*P* for trend
	< 5.7%	**5.7-6.5%**	**≥ 6.5%**		**< 5.5%**	**5.5-6.1%**	**≥ 6.1%**	
Elevated BP	1(ref)	1.55 (0.88-2.74)	17.75** (3.26-96.76)	0.002	1(ref)	1.37 (0.89-2.15)	5.63** (1.88-16.65)	0.006
Dyslipidemia	1	1.53 (0.91-2.57)	5.02* (1.22-20.61)	0.026	1	1.61* (1.10-2.37)	3.34* (1.17-9.54)	0.008
IR	1	2.10** (1.25-3.55）	21.50** (2.56-180.56)	< 0.001	1	2.19** (1.46-3.29)	8.69* (2.54-29.70)	<0.001
MS	1	1.95* (1.04-3.64)	20.80** (4.15-104.22)	< 0.001	1	2.09** (1.27-3.45)	11.63** (3.85-35.10)	< 0.001

Abbreviations: IR: insulin resistance, defined by HOMA-IR > 2.6 and

MS: metabolic syndrome.

* vs ref. $P< 0.05$; ** vs ref. $P < 0.01$.

Figure 1. Comparison between the AUCs of the HbA1c and FBG for IGT (A) and T2DM (B), and the AUC of HbA1c for pre-diabetes (C). FBG means fasting blood glucose. SE means sensitivity. SP means specificity. The green discontinuous line indicates the curve defining the area for the HbA1c, and the blue continuous curve defines the area for FBG. Pre-diabetes was defined either by a FBG ≥5.6 mmol/l (IFG) or 2h-BG ≥7.8mmol/l (IGT).The red arrows indicate the different thresholds (sensitivity, specificity) of HbA1c.

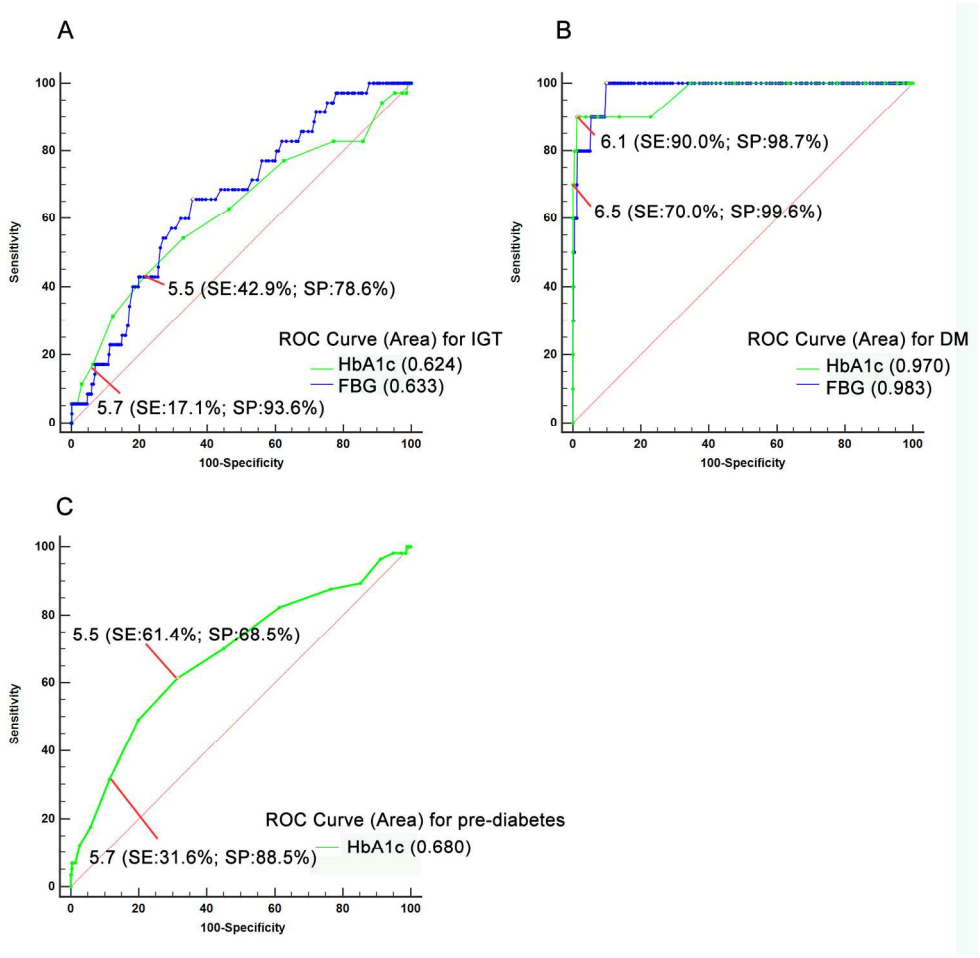

Figure 1. Comparison between the AUCs of the HbA1c and FBG for IGT (A) and T2DM (B), and the AUC of HbA1c for pre-diabetes (C). FBG means fasting blood glucose. SE means sensitivity. SP means specificity. The green discontinuous line indicates the curve defining the area for the HbA1c, and the blue continuous curve defines the area for FBG. Pre-diabetes was defined either by a FBG ≥5.6 mmol/l (IFG) or 2h-BG ≥7.8mmol/l (IGT).The red arrows indicate the different thresholds (sensitivity, specificity) of HbA1c.

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STARD 2015

AIM

STARD stands for "Standards for Reporting Diagnostic accuracy studies". This list of items was developed to contribute to the completeness and transparency of reporting of diagnostic accuracy studies. Authors can use the list to write informative study reports. Editors and peer-reviewers can use it to evaluate whether the information has been included in manuscripts submitted for publication.

EXPLANATION

A **diagnostic accuracy study** evaluates the ability of one or more medical tests to correctly classify study participants as having a **target condition.** This can be a disease, a disease stage, response or benefit from therapy, or an event or condition in the future. A medical test can be an imaging procedure, a laboratory test, elements from history and physical examination, a combination of these, or any other method for collecting information about the current health status of a patient.

The test whose accuracy is evaluated is called **index test.** A study can evaluate the accuracy of one or more index tests. Evaluating the ability of a medical test to correctly classify patients is typically done by comparing the distribution of the index test results with those of the **reference standard**. The reference standard is the best available method for establishing the presence or absence of the target condition. An accuracy study can rely on one or more reference standards.

If test results are categorized as either positive or negative, the cross tabulation of the index test results against those of the reference standard can be used to estimate the **sensitivity** of the index test (the proportion of participants *with* the target condition who have a positive index test), and its **specificity** (the proportion *without* the target condition who have a negative index test). From this cross tabulation (sometimes referred to as the contingency or "2x2" table), several other accuracy statistics can be estimated, such as the positive and negative **predictive values** of the test. Confidence intervals around estimates of accuracy can then be calculated to quantify the statistical **precision** of the measurements.

If the index test results can take more than two values, categorization of test results as positive or negative requires a **test positivity cut-off**. When multiple such cut-offs can be defined, authors can report a receiver operating characteristic (ROC) curve which graphically represents the combination of sensitivity and specificity for each possible test positivity cut-off. The **area under the ROC curve** informs in a single numerical value about the overall diagnostic accuracy of the index test.

The **intended use** of a medical test can be diagnosis, screening, staging, monitoring, surveillance, prediction or prognosis. The **clinical role** of a test explains its position relative to existing tests in the clinical pathway. A replacement test, for example, replaces an existing test. A triage test is used before an existing test; an add-on test is used after an existing test.

Besides diagnostic accuracy, several other outcomes and statistics may be relevant in the evaluation of medical tests. Medical tests can also be used to classify patients for purposes other than diagnosis, such as staging or prognosis. The STARD list was not explicitly developed for these other outcomes, statistics, and study types, although most STARD items would still apply.

DEVELOPMENT

This STARD list was released in 2015. The 30 items were identified by an international expert group of methodologists, researchers, and editors. The guiding principle in the development of STARD was to select items that, when reported, would help readers to judge the potential for bias in the study, to appraise the applicability of the study findings and the validity of conclusions and recommendations. The list represents an update of the first version, which was published in 2003.

More information can be found on http://www.equator-network.org/reporting-guidelines/stard.

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Evaluation of ADA HbA1c Criteria in the Diagnosis of Pre-diabetes and Diabetes in a Population of Chinese Adolescents and Young Adults at High Risk for Diabetes: a Cross-sectional Study

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Evaluation of ADA HbA1c Criteria in the Diagnosis of Pre-diabetes and Diabetes in a Population of Chinese Adolescents and Young Adults at High Risk for Diabetes: a Cross-sectional Study

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Abstract

Objective We aimed to assess HbA1c for the diagnosis of pre-diabetes and diabetes in a population of Chinese youths at risk of metabolic syndrome.

Setting Beijing, China.

Participants A total of 581 subjects aged 14-28 years underwent evaluation including an oral glucose tolerance test (OGTT). Insulin sensitivity, β-cell function and a number of cardiovascular disease risk factors were evaluated. Receiver operating characteristic curves (ROC) were used to assess the screening efficacy of HbA1c.

Results Using OGTT data as a standard, the majority (70.0%, 7/10) of subjects with diabetes would have been diagnosed with HbA1c ≥ 6.5%. In contrast, only 28.1% (16/57) of subjects with pre-diabetes possessed elevated HbA1c's, while the majority (68.4%) had normal HbA1c's. On the contrary, a total of 8.1% (39/479) of youths in the normal HbA1c category (<5.7%) and 21.3% in the pre-diabetes category had pre-diabetes. In the ROC analysis, the area under the curve (AUC) for HbA1c identifying prediabetes, was 0.680 [95%CI 0.640-0.719]; the optimal threshold was 5.5%, with a sensitivity of 61.4% and specificity of 68.5%. For T2DM, the AUC for HbA1c was 0.970 [0.952-0.982], and the optimal threshold was 6.1%, with a sensitivity of 90.0% and a specificity of 98.7%. Applying these new cut-offs, pre-diabetic participants (HbA1c 5.5-6.1%) had lower disposition index and higher risk of dyslipidemia (OR=1.61, [95% CI 1.10-2.37]) and metabolic syndrome (OR=2.09, [1.27-3.45]) than those with normal HbA1c (<5.5%).

Conclusion The ADA's established HbA1c criteria for pre-diabetes and diabetes (5.7% and 6.5%) may not be appropriately applied to adolescents and young adults in China. Our findings suggest that those with HbA1c of 5.5-6.1% already exhibit impaired β-cell function and increased cardio-metabolic risk factors, which may warrant intervention.

Key terms: HbA1c; Diabetes; Pre-diabetes; Metabolic syndrome; adolescents

Strengths and limitations of this study

This study included a well-characterized cohort of adolescents and young adults at risk for diabetes.

All these individuals have undergone an oral glucose tolerance test to evaluate their alterations in insulin sensitivity and β-cell function.

This is the first study in a population of Chinese adolescents and young adults to assess the ADA's HbA1c cutpoints for predicting diabetes or pre-diabetes against the gold-standard OGTT.

However, there was a relatively small sample size of individuals with diabetes by ADA criteria in the population of youth.

While the study cohort was a large population based sample, it may not be representative of the overall Chinese population as we chose to intensely study a subset at risk for the condition of interest (diabetes/pre-diabetes).

Abbreviations

BMI: body mass index; WC: waist circumference; FBG: Fasting blood glucose; SBP: Systolic blood pressure; DBP: Diastolic blood pressure; TG: Triglycerides; TC: Total cholesterol; LDL-C: Low-density lipoprotein cholesterol; HDL□C: High□density lipoprotein cholesterol; CRP: C-reacting protein; OGTT: Oral glucose tolerance test; INS: Insulin; ISI: Insulin sensitivity index; IGI: Insulinogenic index; DIO: Oral disposition index; IFG: Impaired fasting glucose; IGT: Impaired glucose tolerance; HOMA-IR: The index of homeostasis model assessment of insulin resistance; MS: Metabolic syndrome; T2DM: Type 2 diabetes; ROC: Receiver operating characteristic.

Introduction

The incidence of obesity has increased dramatically in recent decades among Chinese children and adolescents. The Global Burden of Disease Study showed that the prevalence of overweight and obesity in children and adolescents in developing countries has increased from 8.4% in 1980 to 13.4% in 2013[1]. With the global surge in obesity, prevalence of diabetes has increased substantially. World Health Organization (WHO) data from 2014 estimated that 347 million people worldwide had diabetes[2]. A nationwide survey conducted by Yang et al. in 2010 showed that the prevalence of diabetes and pre-diabetes among adults in China had reached 9.7% and 15.5%, respectively[3]. As both pre-diabetes and type 2 diabetes (T2DM) have emerged as consequences of childhood obesity[4] the clustering of cardiovascular risk factors in this population[5] heightens concern that obese children and young adults are at risk for complications of diabetes, specifically cardiovascular disease. Thus, early identification of the population predisposed to developing diabetes is critically important if we are to target them for early intervention.

Screening for dysglycemia (diabetes and prediabetes) has traditionally focused on OGTT to identify diabetes, impaired fasting glucose (IFG) and impaired glucose tolerance (IGT). However, due to time, expense, and inconvenience, conducting an oral glucose tolerance test (OGTT) is often not feasible in patient care or population-based studies[6]. Fasting blood glucose (FBG) has been used as an inexpensive alternative to the OGTT, but FBG is also associated with challenges, like the requirement for an 8-h fast. In a study of diabetes screening practices among pediatric clinicians, a strong preference for non-fasting tests was evident[7].

HbA1c has become increasingly popular for diabetes screening among primary care providers due to its many practical advantages including: convenience of sampling, suitability as an index of chronic dysglycemia, low intra-individual variability, and propitious assay standardization[8]. In 2010, the American Diabetes Association (ADA)[9] suggested that HbA1c values of 5.7-6.4% established a diagnosis of

pre-diabetes while a value of ≥6.5% defined diabetes. These recommendations are based on data in adults showing the relationship between HbA1C and the subsequent development of diabetic microvascular complications. However, it remains controversial what HbA1c level should be applied to the definition of pre-diabetes in children and adolescents, with at least three proposed thresholds: 6.0%[10], 5.7%[9]and 5.5%[11]. Furthermore, it is unclear at what ages these HbA1c thresholds should be applied, due to the paucity of longitudinal data in children (and even young adults) which associate these cut points with adverse cardio-metabolic outcomes. Until these long-term outcome data become available, pre-diabetes and diabetes can best be defined by their ability to identify pathophysiologic abnormalities associated with hyperglycemia such as decreased β-cell function and insulin sensitivity[12]. Currently, studies in the Chinese pediatric population are lacking. Therefore, the aim of this study was to assess HbA1c as an instrument to establish the diagnosis of pre-diabetes and diabetes in a population of Chinese adolescents and young adults at increased risk of diabetes.

Materials and methods

Subjects

Subjects were recruited from the cohort of Beijing Children and Adolescents Metabolic Syndrome study (BCAMS). The BCAMS is a longitudinal cohort study of cardiovascular risk factors since childhood. Details of the baseline study have been described previously[13,14]. Briefly, in 2004 a population-based survey was conducted in the Beijing area with a representative sample (n = 19,593, 50% boys) of schoolchildren (aged 6–18 years). Approximately 4500 subjects were identified as being at elevated risk for dysglycemia at baseline due to the presence of one of the following risk factors: overweight defined by body mass index (BMI), total cholesterol (TC) ≥5.2 mmol/L, triglyceride (TG) ≥1.7mmol/L or FBG ≥5.6 mmol/L based on finger capillary blood tests. A follow-up study began in 2012 (8 years after baseline), with subjects recruited

consecutively through various modalities (phone, text and email) for medical examination at Beijing Chaoyang Hospital. A total of 581 subjects who completed medical examination are included in this analysis. Those lost to follow-up were relatively younger and thinner at baseline than those who did follow-up, however, there were no significant difference in gender, pubertal status, blood pressure, fasting TG, low-density lipoprotein cholesterol (LDL-C), high-density lipoprotein cholesterol (HDL-C) and FBG levels (P >0.05). The study was approved by the Ethics Committee at the Beijing Chaoyang Hospital and signed informed consent was obtained from all participants and/or their parents or guardians through all study phases. The BCAMS study was registered at www.clinicaltrials.gov (NCT03421444). Patients or public representatives were not involved at any stage of this study.

Patient and public involvement

No patients were involved in setting the research question or the outcome measures, nor were they involved in the design of this study. No patients were involved in the recruitment to and conduct of the study. There are no plans to disseminate the results of the research to study participants or the relevant patient community.

Clinical measurements

Height, weight, waist circumference (WC) and percent body fat (FAT%) were measured by trained field workers. Participants removed bulky clothing and shoes prior to measurements. Height was measured to the nearest 0.1 cm using a portable stadiometer. WC was measured midway between the lowest rib and the top of the iliac crest. Weight and FAT mass was measured to the nearest 0.1kg using a TANITA Body Composition Analyzer (ModelTBF-300A). Measurements of right arm systolic and diastolic blood pressure (SBP and DBP) were performed 3 times 10 minutes apart and the mean values of the latter two measurements were recorded. BMI was calculated as weight divided by height squared.

Laboratory measurements

Venous blood samples were collected after an overnight (≥ 12h) fast. An OGTT using 75g glucose load was performed with plasma glucose levels in the fasting state (FBG), 0.5-hour (0.5hBG) and 2-hour (2hBG) measured using a hexokinase method. The concentrations of TG, TC and LDL-C were assayed using a standard enzymatic method. HDL-C was assessed with a phosphotungstic acid-Mg method. Serum C-reacting protein (CRP) was measured by immunoturbidimetric assay. Insulin concentrations were measured by monoclonal antibody-based sandwich enzyme-linked immunosorbent assays which was developed in the Key Laboratory of Endocrinology, Peking Union Medical College Hospital. The intra- and inter-assay coefficients of variation (CVs) for insulin were < 5.4% and < 9.0%, respectively, with no cross-reactivity to proinsulin (< 0.05%). HbA1c was assayed using the TOSOH G7 automatic analysis system with high pressure liquid chromatography. This assay is certified by the National Glycohemoglobin Standardization Program (NSPG).

Insulin resistance was estimated by following indices: (1) the homeostasis model assessment of insulin resistance (HOMA-IR) [(fasting insulin mU/L) × (FBG mmol/L)/22.5][15]; (2) Insulin sensitive index (Matsuda Index), [ISI (Matsuda) = 10,000/√((FBG × fasting plasma insulin) × (mean plasma glucose × mean plasma insulin))][16]. Pancreatic β-cell function was assessed by: (1) homeostasis model assessment of β-cell function (HOMA-β) [(20 x fasting insulin)/(FPG-3.5)][17]; (2) insulinogenic index (IGI=ΔInsulin30/ΔGlucose30); (3) the ratio of the total area under the insulin curve to the total area under the glucose curve (total AUC Insulin/Glucose) and (4) the oral disposition index (DI_O =IGI×ISI), which is the product of insulin sensitivity and insulin secretion [18,19].

Definitions

Dysglycemia (IFG, IGT, prediabetes, diabetes) was defined according to current American Diabetes Association guidelines[9]. Metabolic syndrome (MS) was diagnosed according to 2009 Joint Task Force

harmonization criteria, with subjects exhibiting at least three of the following five components [20]: (1) central obesity: WC ≥ 90th percentile for age and sex in 10 -16 years, or ≥ 90 cm for male and ≥ 80 cm for female; (2) IFG, IGT or diabetes; (3) BP: ≥130/85 mmHg; (4) HDL-C < 1.03mmol/L in males, < 1.29 mmol/L in females and (5) TG ≥ 1.70mmol/L. According to Chinese age- and sex-specific BMI cutoffs[21], adolescents were classified as overweight if BMI was between the 85th and 95th percentile, and obese if BMI was above 95th percentile. Subjects older than 18 year-old were classified overweight if BMI ≥ 24 kg/m^2, or obese if BMI ≥ 28 kg/m^2.

Data analysis

All statistical analyses were carried out using the Statistical Package for Social Sciences (SPSS 19.0 for windows). Continuous variables were tested for normality using a Kolmogorov-Smirnov test. Non-normal distribution values used in the analyses were log-transformed to improve normality. Results are expressed as mean ± standard deviation (SD). Group comparisons across three HbA1c categories were made with ANOVA with Bonferroni *post hoc* comparison test. Agreement between HbA1c, fasting glucose category and OGTT 2-h glucose was also assessed. K coefficients were reported. Receiver operating characteristic (ROC) curve analysis was performed for HbA1c and FBG to discriminate pre-diabetes from normal glucose tolerance (NGT) and T2DM, from NGT and IGT using a logistic procedure. Area under the ROC curve (AUC) was considered as an effective measure of inherent validity of a diagnostic test. The mean values of variables were studied by analysis of variance. Multivariate logistic regression models were used to estimate ORs for IR, MS and its components. Level of significance was accepted as $P < 0.05$.

Results

Subjects characteristics

The mean age of the entire population was 20.2 ± 2.9 years (female 46.8%). The prevalence rates of

obesity, high blood pressure, dyslipidemia and MS were 32.6%, 20.2%, 29.5% and 14.5%, respectively. Of 581 subjects, 18 refused to conduct 2h-OGTT. Using the HbA1c criteria recommended by ADA, the prevalence of T2DM was 1.5% (9/581) and pre-diabetes was 13.4% (78/581), whereas employing OGTT criteria yielded somewhat different prevalence rates [IFG 4.8% (28/581), IGT 6.2% (35/563), IFG and /or IGT 10.1% (57/563) and T2DM 1.7% (10/581)].

Comparisons between HbA1c and fasting glucose

The average HbA1c level was 5.4 ± 0.6%. HbA1c showed a strong positive correlation to FBG (r = 0.734, $P < 0.001$), 2h-BG (r = 0.694, $P < 0.001$), but a modest negative correlation with ISI (r = -0.177, $P<0.001$), IGI (r = -0.258, $P < 0.001$) and DIO (r = -0.389, $P < 0.001$) (Table 1). There were also modest correlations between HbA1c and various cardio-metabolic parameters, such as TG (r = 0.159, $P < 0.001$), TC (r = 0.157, $P < 0.001$), LDL-C (r = 0.176, $P < 0.001$), HDL-C (r = -0.103 $P < 0.05$), SBP (r = 0.143, $P = 0.001$) and hsCRP (r = 0.111, $P < 0.05$). FBG showed similar correlation to these cardio-metabolic parameters, except for hsCRP ($P = 0.125$).

The classification of subjects using HbA1c versus OGTT is shown in Table 2. First, using OGTT data as a standard, the majority (7/10, 70.0%) of subjects with diabetes would have been diagnosed by HbA1c ≥ 6.5%. In contrast, only 25.7% (9/35) of subjects with IGT possessed elevated HbA1c's indicative of pre-diabetes, while the majority (68.6%) had normal HbA1c's. Second, the majority (87.6%) of the subjects with NGT would be identified with HbA1c<5.7%, while 12.4 % were classified with pre-diabetes or diabetes. On the other hand, of those considered to have diabetes by 2h-OGTT criteria, 3 of 10 (30.0 %) were missed by HbA1c, while those identified as pre-diabetic on an OGTT (i.e., IFG and/or IGT), 39 of 57 (68.4 %) were missed by HbA1c criteria.

However, of those diagnosed with diabetes using OGTT, only 4 of 10 (40.0%) were identified by their FBG values, and among those with IGT, only 2 of 35 (5.7%) were identified with IFG. So, using FPG

criteria to identify dysglycemia would miss the majority of IGT and T2DM and using HbA1c was only moderately better (as demonstrated by the low k coefficients between either FBG (k=0.16) or HbA1c (k= 0.21) and OGTT.

ROC curve analysis

Figures 1A and 1B represent the diagnostic accuracy of the HbA1c and FBG, for IGT and diabetes identified by OGTT, respectively. The AUC for HbA1c was 0.624 [95%CI 0.582-0.664] and the AUC for FBG was 0.663[0.576-0.749]. The optimal HbA1c threshold for identifying IGT was 5.5%, with a sensitivity of 42.9% and specificity of 78.6%. To identify diabetes, the AUC for HbA1c was 0.970 [0.952-0.982], and for IFG the AUC was 0.789 [0.706-0.872]. The optimal HbA1c threshold of 6.1%, identified diabetes with 90.0% sensitivity and 98.7% specificity.

In light of the inconsistency between IFG and IGT when identifying dysglycemia, we defined pre-diabetes as either IFG or IGT, and evaluated HbA1c test performance with ROC. As shown in Figure 1C and Table 3, the AUC of HbA1c for pre-diabetes was 0.680 (95%CI 0.640-0.719), and the optimal threshold of HbA1c was still 5.5%, with a sensitivity of 61.4% and specificity of 68.5%. Moreover, as shown in Figure 1and Table 3, compared with the ADA criteria for HbA1c (5.7%), lowering the HbA1c threshold to 5.5% doubles the sensitivity of this test, while only moderately affecting the specificity.

Comparisons of metabolic characteristics according to different HbA1c criteria

To compare the segregation of metabolic characteristics among groups identified by our proposed HbA1c thresholds versus the ADA criteria, we stratified the population according to HbA1c categories (Table 4). Age distribution, BMI, WC, DBP, FBG and 2h-BG (all $P < 0.05$) were all different among the three categories defined by either ADA or our proposed thresholds, while HDL-C was not. Not surprisingly, there were more subjects (32.9% vs. 13.4%) classified as pre-diabetic based on HbA1c 5.5-6.1% than by ADA criteria of 5.7-6.4%. Similarly, a greater number of subjects 16 (2.8%) vs. 9 (1.5%) would be considered to

have T2DM by our criteria.

Table 4 compares the ability of these differing HbA1c strata to delineate levels of β-cell dysfunction and insulin resistance derived from FPG and OGTT measures. Subjects in the HbA1c 5.5-6.1% vs <5.5% categories demonstrated no difference with regard to insulin resistance indices (HOMA-IR and ISI), and neither did those with HbA1c 5.7-6.4% comparing with those < 5.7%. However, subjects with HbA1c 5.5-6.1% showed a significantly lower IGI and DI_O compared to those with HbA1c < 5.5%. Notably, these differences were not as pronounced among groups classified using ADA criteria, especially with regard to IGI.

Regardless of the HbA1c thresholds employed, lipids measures (TC, TG and LDL-C) were significantly higher in diabetes than in normal or pre-diabetes categories, whereas hsCRP was highest in the pre-diabetic individuals ($P < 0.05$). Moreover, high HbA1c was associated with a higher prevalence of obesity, hypertension, IR and dyslipidemia. Of those with HbA1c 5.5-6.1%, 18.8 % had MS, compared with only 10.5 % of those with HbA1c < 5.5%. Applying our HbA1c thresholds (Table 5), the odds ratios for dyslipidemia, insulin resistance and MS in pre-diabetic versus non-diabetic individuals were 1.61, 2.19 and 2.09 respectively, which is somewhat higher than if ADA criteria were employed.

Discussion

This cross sectional study demonstrates that the ADA's HbA1c thresholds of 5.7% and 6.5% had low sensitivity for classifying pre-diabetes (31.6%) and diabetes (63.6%) as defined by OGTT in a young Chinese population. Rather, we propose a threshold of 6.1% for identifying diabetes, with a sensitivity of 81.8% and specificity of 98.8%, and 5.5% for detecting pre-diabetes, with a sensitivity of 68.5% and specificity of 61.4%. We observed that the use of HbA1c thresholds of 5.5% and 6.1% would significantly improve the sensitivity of these measures without adversely affecting their specificities. Our data are in

agreement with reports in children[22-25] which have concluded that HbA1c thresholds of 5.7-6.4% and ≥6.5% are insufficient to detect pre-diabetes and diabetes.

Several studies have assessed HbA1c cut points for predicting diabetes or pre-diabetes against the gold-standard OGTT and concluded that HbA1c is a poor predictor of pre-diabetes and T2DM in young people. Lee et al.[23] examined the ability of various tests to diagnosis pre-diabetes and diabetes in obese children; comparing HbA1c, fructosamine and random glucose with OGTT. They found that all of these tests were poor discriminators, and led to missed cases of dysglycemia in children. Similarly, Nowicka et al.[24] suggested that an HbA1c of 5.8% for identifying T2DM in a multiethnic cohort of 1,156 obese children and adolescents from the United States. Because of the lower prevalence of diabetes in pediatric as opposed to adult populations, the utility of A1c for detecting diabetes may be suspect, and at the very least, thresholds require adjustment for the population under study. For this reason, Kester, et al [25] suggested caution when adopting HbA1c as a principal diagnostic method in children, and called for prospective studies of pre-diabetes and T2DM in obese pediatric populations to determine HbA1c cutoff points.

FBG has been used as an inexpensive alternative to OGTT, especially when screening for MS. In our study, we compared HbA1C versus FBG to detect dysglycemia. In the subjects categorized as pre-diabetic by OGTT, 31.6% showed laboratory evidence of being at risk for DM on the basis of HbA1c, while only 5 (6.2%) were identified with IFG. Of the 10 classified with DM by OGTT, 7 subjects were detected with HbA1c, while only 4 would be identified with DM on the basis of FBG. These findings are consistent with Chan et al who showed that FPG performed poorly in obese 10-18 year olds compared to HbA1c and OGTT when identifying dysglycemia which was detected using a blinded continuous glucose monitoring (CGM) device[26]. Furthermore, hemoglobin A1c may present certain advantages over other tests. For example, the multiethnic Healthy Study cohort demonstrated greater consistency of HbA1c versus FBG in a prospective trial in middle school children[27].

The debate over which test (HbA1c, FBG or 2h-OGTT) is best to identify relevant glycemic abnormalities in youth will ultimately require decades of prospective study to determine which test is most prognostic of cardiovascular and microvascular consequences. Until these long-term outcome studies become available, pre-diabetes and diabetes can be defined alternately by pathophysiologic abnormalities associated with diabetes such as declining insulin sensitivity and β-cell function. Thus, the present study compares the ADA criteria with our proposed cutoff points to detect those alterations in insulin sensitivity and β-cell function based on OGTT. We demonstrate that an HbA1c in the range of 5.5-6.1% is associated with reduced β-cell function (IGI, HOMA-β) as well as DI_O, which is an established metabolic predictor of progression to diabetes[28]. We found progressively declining DI_O across the continuum of HbA1c from < 5.5% to 5.5-6.1% to > 6.1%. In contrast, the established ADA HbA1c criteria did not detect a difference in β-cell function (IGI) as HbA1c progressed from < 5.7% to 5.5-6.4%. This implies that our proposed thresholds are more rational for defining diabetes risk, at least in a young Chinese population.

Studies in adults and children have shown that prediabetes is associated with increased prevalence of cardiovascular risk factors[5,29]. We also found a clustering of cardiovascular risk factors among subjects at risk for diabetes defined by our HbA1c thresholds. In the HbA1c 5.5-6.1%, compared with those < 5.5%, elevated HbA1c was associated with known risk factors for cardiovascular disease, including waist circumference, DBP, TC, TG and LDL-C, hsCRP as well as a more than a twofold increased risk of having IR. Thus, our findings are consistent with an evolving consensus that, HbA1c may identify a population with increased risk of microvascular and macrovascular complications.

Cardiovascular disease (CVD) generally presents during adulthood, but the antecedents of this adult disease may be detectable in childhood. Elevated lipid and blood pressure (BP) levels have been associated with an increased risk of CVD, and these risk factors track from childhood into adulthood [30,31] Although this has not been definitively demonstrated in a prospective study of dysglycemia which spans from childhood

into adult life, it is reasonable to conclude that a similar persistence, if not a progression, of glycemic abnormalities would be observed. Adolescents with pre-diabetes or T2DM face many years of hyperglycemia and cardiovascular disease, and thus, may have an increased lifetime risk of developing complications. In fact, a large proportion of American adolescents have microalbuminuria and cardiovascular risk factors at diagnosis of T2DM[32].

This study has several strengths and weaknesses. Considerable strength is drawn from the well-characterized cohort of adolescents and young adults at risk for diabetes. However, relatively few from this cohort had diabetes by ADA criteria. In addition, pubertal stage, which can influence insulin sensitivity and lipid levels, was not evaluated in this study. However, the small proportion of participants (<12%) less than 16 years old suggests that the vast majority of participant were post-pubertal, thus rendering this as only a minor concern. Furthermore, since puberty is associated with age, and we did include age as a covariate when comparing clinical features across HbA1c categories, there was some adjustment for the effect of pubertal stage. Finally, compared with our original population at baseline, the follow-up group is relatively small, which may introduce the potential for bias. Nonetheless, there were no significant difference in gender, pubertal status, or major cardio-metabolic parameters at baseline between those who followed-up versus those lost to follow-up.

In conclusion, the ADA's established HbA1c criteria for pre-diabetes and diabetes (5.7% and 6.5%) may not be appropriately applied to adolescents and young adults, especially in the Chinese population. Our findings suggest that those with HbA1c of 5.5 - 6.1% already exhibit impaired β-cell function and increased cardio-metabolic risk, which may warrant intervention.

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Contributors GL analyzed data and drafted the manuscript; LWH, YHW, JLF and YL contributed to data collection; YLZ contributed to the data analysis and revised the manuscript; SMW was responsible for the concept, and design of the study and contributed to the data interpretation and reviewed/edited the manuscript. ML contributed to the concept, design of the study, analyzed the data and revised the manuscript. SG was responsible for the concept, design, and data collection in the BCAMS follow-up study, and contributed to acquisition and interpretation of the data, and revised the manuscript.

Sharing statement Additional details on data presented in the current study are available by emailing liming@pumch.cn or gaoshanmw@163.com.

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Competing interests None declared.

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Table 1. Correlation of HbA1c, glucose with cardio-metabolic risk parameters

	HbA1c	**FBG**	**2h-BG**
HbA1c (%)	1	0.734**	0. 694**
FBG (mmol/l)	0.734**	1	0.718**
2h-BG (mmol/l)	0. 694**	0.718**	1
TG (mmol/l)	0.159**	0.182**	0.196**
LDL-C (mmol/l)	0.176**	0.108**	0.152**
TC (mmol/l)	0.157**	0.032	0.048
HDL-C (mmol/l)	-0.103*	-0.095*	-0.102*
SBP (mmHg)	0.143**	0.151**	0.219**
DBP (mmHg)	0.209**	0.63**	0.238**
MS score	0.270**	0.215**	0.326**
Ln CRP (mg/l)[#]	0.112*	0.069	0.126**
Ln ISI	-0. 177**	-0.226**	-0.304**
Ln IGI	-0.258**	-0.213**	-0.282**
Ln DIO	-0.389**	-0.386**	-0.528**
Ln FINS (mU/L)	0.169**	0.182**	0.198**
Ln 0.5h-INS (mU/L)	-0.096*	-0.121**	-0.083
Ln 2h-INS (mU/L)	0.038	-0.025	0.357**

Abbreviations: FBG: Fasting blood glucose; TG: Triglycerides; TC: Total cholesterol; LDL‐C: Low‐density lipoprotein cholesterol; HDL‐C: High‐density lipoprotein cholesterol; SBP: Systolic blood pressure; DBP: Diastolic blood pressure; CRP: C-reacting protein; INS: insulin; ISI: insulin sensitivity index; IGI: insulinogenic index; DIO: oral disposition index and MS metabolic syndrome.

MS score: numbers of MS components

* $P < 0.05$; ** $P < 0.01$.

Table 2. The frequency of subjects with prediabetes and T2DM meeting the diagnostic criteria (HbA1c, FBG and 2h-BG after 75 g-OGTT)

OGTT	HbA1c *			Total	FBG #			Total
	NGT (< 5.7%)	At risk for diabetes (5.7-6.4%)	T2DM (> 6.4%)		NGT (< 5.6 mmol/l)	IFG (5.6-7.0 mmol/l)	T2DM (> 7.0 mmol/l)	
NGT	454 (87.6)	64 (12.4)	0	518	497 (95.9)	21 (4.1)	0	518
IGT	24(68.6)	9 (25.7)	2 (5.7)	35	33 (94.3)	2 (5.7)	0	35
T2DM	1(10.0)	2(20.0)	7 (70.0)	10	2(20.0)	4 (40.0)	4 (40.0)	10
Total	479 (85.1)	75 (13.3)	9 (1.6)	563@	532(94.5)	27 (4.8)	5 (0.7)	563@

Abbreviations: FBG: Fasting blood glucose; NGT: Normal glucose tolerance; IFG: Impaired fasting glucose; IGT: Impaired glucose tolerance and T2DM: Type 2 diabetes.

Numbers in brackets are percentages of horizontal total

*kappa coefficient 0.21; # kappa coefficient 0.16; @ 18 of 581 subjects disagreed to undergo 2h-OGTT.

Table 3. Test performance characteristics of specific HbA1c thresholds for detecting prediabetes and according to OGTT

HbA1c Threshold	Prediabetes (IFG + IGT)			T2DM		
	sensitivity	1-specificity	Sensitivity + specificity	sensitivity	1-specificity	Sensitivity + specificity
2.7	1.000	1.000	1.000	1.000	1.000	1.000
3.8	1.000	0.998	1.002	1.000	0.998	1.002
4.0	1.000	0.996	1.004	1.000	0.996	1.004
4.1	1.000	0.994	1.006	1.000	0.995	1.005
4.3	1.000	0.992	1.008	1.000	0.993	1.007
4.5	1.000	0.990	1.010	1.000	0.991	1.009
4.7	0.982	0.986	0.997	1.000	0.986	1.014
4.8	0.982	0.972	1.011	1.000	0.973	1.027
4.9	0.982	0.950	1.033	1.000	0.953	1.047
5.0	0.965	0.911	1.054	1.000	0.917	1.083
5.1	0.895	0.853	1.042	1.000	0.857	1.143
5.2	0.877	0.764	1.113	1.000	0.776	1.224
5.3	0.825	0.615	1.210	1.000	0.635	1.365
5.4	0.702	0.452	1.250	1.000	0.476	1.524
5.5	0.614	0.315	1.300	1.000	0.344	1.656
5.6	0.491	0.200	1.292	0.900	0.228	1.672
5.7	0.316	0.115	1.201	0.900	0.136	1.764
5.8	0.175	0.058	1.117	0.900	0.071	1.829
5.9	0.123	0.026	1.097	0.900	0.036	1.864
6.0	0.070	0.014	1.056	0.900	0.020	1.880
6.1	0.070	0.006	1.064	0.900	0.013	1.887
6.2	0.070	0.004	1.066	0.800	0.011	1.789
6.3	0.053	0.004	1.049	0.800	0.009	1.791
6.4	0.035	0.002	1.033	0.800	0.005	1.795
6.5	0.035	0.000	1.035	0.700	0.004	1.696
6.8				0.700	0.000	1.700
7.1				0.600	0.000	1.600
7.4				0.500	0.000	1.500
8.4				0.400	0.000	1.400
9.5				0.300	0.000	1.300
10.4				0.200	0.000	1.200
12.5				0.100	0.000	1.100
14.8				0.000	0.000	1.000

Abbreviations: IFG: impaired fasting glucose; IGT: impaired glucose tolerance and T2DM: type 2 diabetes.

Table 4. Clinical features of the study population according to HbA1c categories

	ADA criteria			*P*	Our proposed criteria			*P*
	< 5.7%	**5.7-6.4%**	**≥ 6.5%**		**< 5.5%**	**5.5-6.1%**	**≥ 6.1%**	
N	494	78	9		374	191	16	
Age (years)	20.30 (0.13)	19.28 (0.33)†	22.11(0.86)§	0.001	20.38 (0.14)	19.67 (0.23)†	21.33 (0.90)	0.010
Sex (M/F)	253/241	49/29	6/3	0.115	189/185	108/83	11/5	0.176
BMI (kg/m^2)	25.41 (0.24)	27.53 (0.83)†	28.73 (2.82)	<0.001	25.03(0.26)	26.90(6.46)†	28.65(2.11)§	< 0.001
WC (cm)	84.4 (0.6)	89.7 (2.1)†	97.7 (7.8)§	<0.001	83.4 (0.7)	88.5 (1.12)†	94.0(5.6)§	< 0.001
SBP (mmHg) *	114.7(0.5)	114.5 (1.2)	127.5(3.6) §¶	0.002	114.6(0.6)	115.1 (0.8)	119.2 (2.7)	0.251
DBP (mmHg) *	72.9(0.39)	75.2(1.0)	81.7(2.9)§	0.003	72.3 (0.5)	74.9 (0.6) ‡	78.3 (2.2)‖	<0.001
FBG (mmol/l)*	4.85 (0.03)	4.99 (0.08)	9.24(0.23)§¶	< 0.001	4.82 (0.04)	4.94(0.06)	7.61 (0.19)‖&	< 0.001
2h-BG (mmol/l)*	5.87 (0.07)	6.46(0.18)†	16.43(0.51)§¶	< 0.001	5.80 (0.09)	6.15 (0.12)	12.89 (0.41)‖&	< 0.001
IFG, n (%)	16(3.2%)	8(10.3%)†	4 (44.4%)§¶	< 0.001	6 (1.6%)	15(7.9%))‡	7 (43.8%)‖&	< 0.001
IGT, n (%)	24(5.0%)	9 (12.0%)†	2 (22.2%)§¶	< 0.001	16(4.4%)	17 (9.2%))‡	2 (12.5%)‖&	< 0.001
Pre-diabetes, n (%)	39(8.1%)	16(21.3%)†	2(22.2%)§	< 0.001	22 (6.1%)	30(16.3%)‡	4 (25.0%)‖	< 0.001
T2DM, n (%)	1(0.2%)	2 (2.6%)	7(77.8%)§¶	< 0.001	0(0%)	1(0.5%)	9 (56.3%)‖&	< 0.001
Ln FINS (mIU/L)#	1.92(0.03)	1.97(0.07)	2.64(0.21)§¶	0.003	1.92(0.03)	1.95(0.04)	2.13(0.16)	0.421
Ln 2h-INS (mU/L)#	3.58(0.03)	3.76(0.09)	3.73(0.30)	0.164	3.60(0.04)	3.60(0.06)	3.76(0.21)	0.778
Ln HOMA-IR#	0.38(0.02)	0.46(0.07)	1.58(0.22)§¶	< 0.001	0.38(0.03)	0.42(0.05)	0.91(0.17)‖&	0.007
Ln HOMA-β#	4.66(0.03)	4.63(0.07)	4.30(0.22)	0.280	4.68(0.03)	4.64(0.04)	4.09(0.16)‖&	0.002

Ln ISI[#]	1.81(0.02)	1.72(0.06)	0.95(0.21)§¶	< 0.001	1.81(0.03)	1.79(0.04)	1.45(0.15)‖&	0.068
Ln IGI[#]	0.25(0.04)	0.12(0.09)	-1.20(0.3)§¶	< 0.001	0.30(0.04)	0.14(0.06)‡	-1.14(0.22)‖&	< 0.001
Ln DIO[#]	2.06(0.03)	1.84(0.09)†	-0.21(0.32)§¶	< 0.001	2.11(0.04)	1.92(0.06)‡	0.38(0.21)‖&	< 0.001
LDL-C (mmol/l)*	2.50(0.03)	2.62(0.08)	3.47(0.24)§¶	< 0.001	2.49(0.04)	2.58(0.05)	2.95(0.18)‖	0.022
HDL-C (mmol/l)*	1.43(0.01)	1.45 (0.03)	1.35 (0.10)	0.609	1.44(0.02)	1.42(0.02)	1.50(0.07)	0.576
TC (mmol/l)*	4.30(0.04)	4.56(0.10)†	5.55(0.33)§¶	< 0.001	4.30(0.05)	4.40(0.07)	5.12(0.24)‖&	0.003
TG (mmol/l)*	1.10(0.04)	1.29 (0.09)	2.24 (0.26)§¶	<0.001	1.08(0.04)	1.12(0.06)	2.30(0.20)‖&	< 0.001
Ln-CRP (mg/l)[#]	0.09(0.05)	0.45 (0.14)†	-0.03 (0.39)	0.043	0.015(0.061)	0.34(0.084)‡	0.283(0.302)	0.008
Obesity, n (%)	152(30.8%)	32 (41.0%)	5 (55.6%)	0.134	108(29.0%)	72(37.7%)	9(56.3%)	0.024
MS, n (%)	61(12.4%)	16(20.8%)	7 (77.8%)§¶	< 0.001	39(10.5%)	36(18.8%)‡	9(60.0%)‖&	< 0.001

Abbreviations: BMI: Body mass index; WC: Waist circumference; SBP: Systolic blood pressure; DBP: Diastolic blood pressure; FBG: Fasting blood glucose; IFG: impaired fasting glucose; IGT: impaired glucose tolerance; T2DM: type 2 diabetes; FINS: fasting insulin; HOMA-IR: the index of homeostasis model assessment of insulin resistance; ISI: insulin sensitivity index; IGI: insulinogenic index; DIO: oral disposition index; TG: Triglycerides; TC: Total cholesterol; LDL‐C: Low‐density lipoprotein cholesterol; HDL‐C: High‐density lipoprotein cholesterol; CRP: C-reacting protein and MS: metabolic syndrome.

*adjusted for age, sex and BMI; [#] Log transformed and adjust for age, sex and BMI; Data were shown as mean (SE) or number (percentage).

†: < 5.7% vs. 5.7-6.4%; §: < 5.7% vs. > 6.4%; ¶: 5.7%-6.4% vs. > 6.4%;

‡: < 5.5% vs. 5.5-6.1%; ‖: < 5.5% vs. > 6.1%; & : 5.5-6.1% vs. > 6.1%.

Table 5. Age and sex adjusted ORs and 95% CIs for IR, MS and its components according to HbA1c categories

	ADA Criteria			*p* for trend	Our proposed criteria			*P* for trend
	< 5.7%	**5.7-6.5%**	**≥ 6.5%**		**< 5.5%**	**5.5-6.1%**	**≥ 6.1%**	
Elevated BP	1(ref)	1.55 (0.88-2.74)	17.75** (3.26-96.76)	0.002	1(ref)	1.37 (0.89-2.15)	5.63** (1.88-16.65)	0.006
Dyslipidemia	1	1.53 (0.91-2.57)	5.02* (1.22-20.61)	0.026	1	1.61* (1.10-2.37)	3.34* (1.17-9.54)	0.008
IR	1	2.10** (1.25-3.55）	21.50** (2.56-180.56)	< 0.001	1	2.19** (1.46-3.29)	8.69* (2.54-29.70)	<0.001
MS	1	1.95* (1.04-3.64)	20.80** (4.15-104.22)	< 0.001	1	2.09** (1.27-3.45)	11.63** (3.85-35.10)	< 0.001

Abbreviations: IR: insulin resistance, defined by HOMA-IR > 2.6 and

MS: metabolic syndrome.

* vs ref. $P< 0.05$; ** vs ref. $P < 0.01$.

Figure 1. Comparison between the AUCs of the HbA1c and FBG for IGT (A) and T2DM (B), and the AUC of HbA1c for pre-diabetes (C). FBG means fasting blood glucose. SE means sensitivity. SP means specificity. The green discontinuous line indicates the curve defining the area for the HbA1c, and the blue continuous curve defines the area for FBG. Pre-diabetes was defined either by a FBG ≥5.6 mmol/l (IFG) or 2h-BG ≥7.8mmol/l (IGT).The red arrows indicate the different thresholds (sensitivity, specificity) of HbA1c.

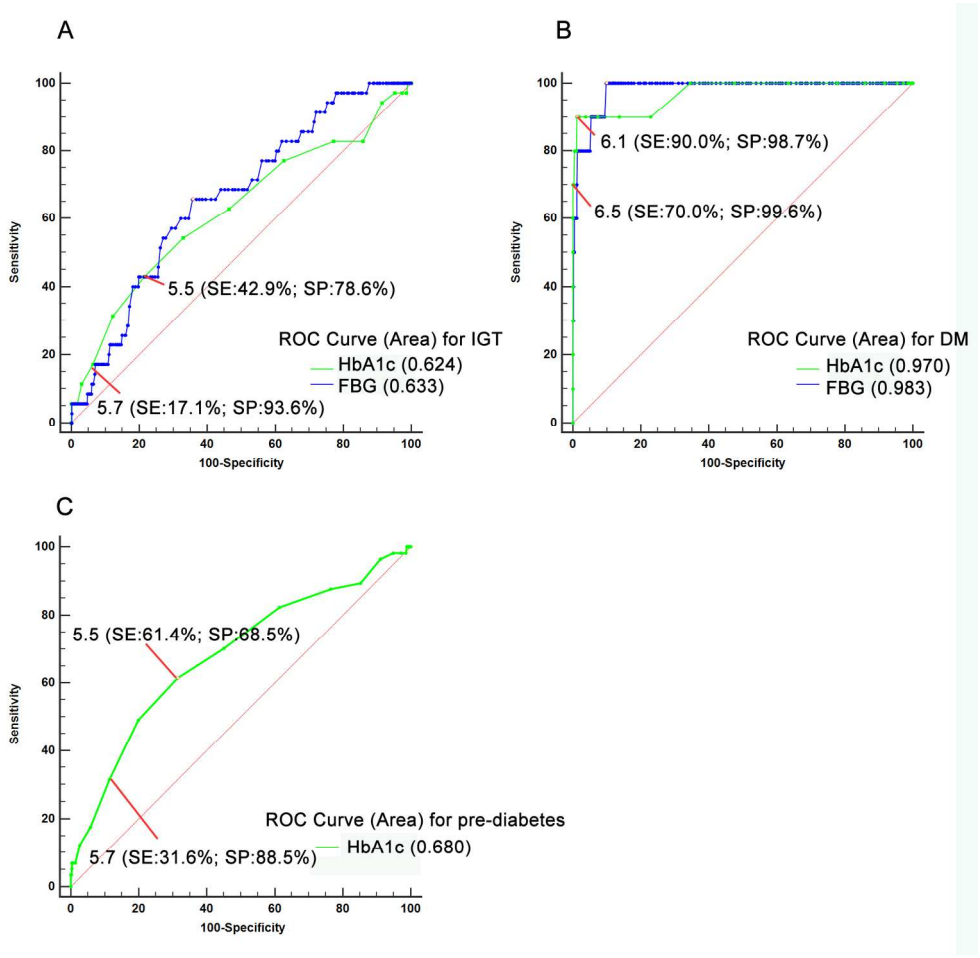

Figure 1. Comparison between the AUCs of the HbA1c and FBG for IGT (A) and T2DM (B), and the AUC of HbA1c for pre-diabetes (C). FBG means fasting blood glucose. SE means sensitivity. SP means specificity. The green discontinuous line indicates the curve defining the area for the HbA1c, and the blue continuous curve defines the area for FBG. Pre-diabetes was defined either by a FBG ≥5.6 mmol/l (IFG) or 2h-BG ≥7.8mmol/l (IGT).The red arrows indicate the different thresholds (sensitivity, specificity) of HbA1c.

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STARD 2015

AIM

STARD stands for "Standards for Reporting Diagnostic accuracy studies". This list of items was developed to contribute to the completeness and transparency of reporting of diagnostic accuracy studies. Authors can use the list to write informative study reports. Editors and peer-reviewers can use it to evaluate whether the information has been included in manuscripts submitted for publication.

EXPLANATION

A **diagnostic accuracy study** evaluates the ability of one or more medical tests to correctly classify study participants as having a **target condition.** This can be a disease, a disease stage, response or benefit from therapy, or an event or condition in the future. A medical test can be an imaging procedure, a laboratory test, elements from history and physical examination, a combination of these, or any other method for collecting information about the current health status of a patient.

The test whose accuracy is evaluated is called **index test.** A study can evaluate the accuracy of one or more index tests. Evaluating the ability of a medical test to correctly classify patients is typically done by comparing the distribution of the index test results with those of the **reference standard**. The reference standard is the best available method for establishing the presence or absence of the target condition. An accuracy study can rely on one or more reference standards.

If test results are categorized as either positive or negative, the cross tabulation of the index test results against those of the reference standard can be used to estimate the **sensitivity** of the index test (the proportion of participants *with* the target condition who have a positive index test), and its **specificity** (the proportion *without* the target condition who have a negative index test). From this cross tabulation (sometimes referred to as the contingency or "2x2" table), several other accuracy statistics can be estimated, such as the positive and negative **predictive values** of the test. Confidence intervals around estimates of accuracy can then be calculated to quantify the statistical **precision** of the measurements.

If the index test results can take more than two values, categorization of test results as positive or negative requires a **test positivity cut-off**. When multiple such cut-offs can be defined, authors can report a receiver operating characteristic (ROC) curve which graphically represents the combination of sensitivity and specificity for each possible test positivity cut-off. The **area under the ROC curve** informs in a single numerical value about the overall diagnostic accuracy of the index test.

The **intended use** of a medical test can be diagnosis, screening, staging, monitoring, surveillance, prediction or prognosis. The **clinical role** of a test explains its position relative to existing tests in the clinical pathway. A replacement test, for example, replaces an existing test. A triage test is used before an existing test; an add-on test is used after an existing test.

Besides diagnostic accuracy, several other outcomes and statistics may be relevant in the evaluation of medical tests. Medical tests can also be used to classify patients for purposes other than diagnosis, such as staging or prognosis. The STARD list was not explicitly developed for these other outcomes, statistics, and study types, although most STARD items would still apply.

DEVELOPMENT

This STARD list was released in 2015. The 30 items were identified by an international expert group of methodologists, researchers, and editors. The guiding principle in the development of STARD was to select items that, when reported, would help readers to judge the potential for bias in the study, to appraise the applicability of the study findings and the validity of conclusions and recommendations. The list represents an update of the first version, which was published in 2003.

More information can be found on http://www.equator-network.org/reporting-guidelines/stard.