GLYCATED HEMOGLOBIN AND PREDIABETES:
A SYSTEMATIC REVIEW OF AN EMPIRICAL EPIDEMIOLOGIC APPROACH TO DIABETES PREVENTION

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Abstract

Background: Currently, there is disagreement on the minimum threshold of HbA1C to identify adults at risk of developing T2DM. Among highly developed countries, some propose 6.0% as research showed that at levels of 6.0% or higher, there is the potential to suffer from diabetes-related retinopathy (Tapp et al. 2008). Some others, use 5.7%, based on statistical prediction models demonstrating this as the level at which relative and absolute risks to develop T2DM in the long term starts (ADA 2019, Di Pino et al. 2017).

Methods: Three electronic databases, PUBMED, EMBASE and COCHRANE, were searched according to a pre-determined protocol published on PROSPERO (ID CRD42019134344). A search of the grey literature was also conducted. A total of three reviewers were employed: two independent reviewers DK and TK, and a third one MO, to resolve any possible dispute between the first two. The GRADEpro online free version software approach was utilized to assess the quality of the selected literature and the data contained in it, integrating in the tables the quality analysis from CASP. Data extraction was done by Word Document, while Zotero 5.0.60 was employed to keep account of all references. Finally, due to the heterogeneity of the methods in the selected literature, it proved impossible to attempt any aggregation of data. As a result, a narrative synthesis was elected as the preferred method to analyze and report on the overall results.

Findings: Three out of four total studies suggest that an HbA1C of 6.0-6.4% is highly predictive of future T2DM and that the chances of reversion are improbable. One of these suggests that 5.7-6.4% is the most likely range to yield 50% chances of future normoglycemia. The remaining two suggest 5.5% identifies pre-diabetics with the highest chances of. One of these two studies agrees that according to the risk factors from CDC and Diabetes Canada, the preferred minimum cut-off level for HbA1C should be 5.7%. The fourth paper suggests that 6.0% is the ideal minimal cut-off point but it is the weakest study in the review due to considerable bias.

Conclusion: The data relatively suggests 5.7-6.4% as the ideal HbA1C range for identifying pre-diabetic adults. Instead, 6.0-6.4% rather helps to predict an almost sure future development of T2DM, precluding any hope for reversion. Given the moderate to very-low quality of the data, however, the evidence produced cannot be considered indisputable. A HbA1C of 6.0% as a minimum threshold, nonetheless, appears to be no longer viable.

Background

One of the key markers of Type-2 Diabetes Mellitus (T2DM) is the elevated presence of Glycated Hemoglobin A or "HbA1C". The established approach to T2DM prevention is to predict the future development of T2DM in those at increased risk (pre-diabetics) as indicated

by moderate levels of HbA1C. At the moment, there is disagreement on the critical threshold of HbA1C to identify adults at risk. Among highly developed countries, some like Canada and the United Kingdom follow the guidelines offered by the World Health Organization (WHO 2011), which propose 6.0% as the minimum cutoff point to identify pre-diabetes. This value is based on research showing that at levels of 6.0% or higher, there is the potential to suffer from diabetes-related retinopathy (Tapp et al. 2008). Other countries, such as the USA and Italy and Israel, use 5.7% as the critical cut-off, based on statistical prediction models demonstrating that this is the level at which relative and absolute risks to develop T2DM in the long term starts (ADA 2019, Di Pino et al. 2017). According to a publication from the Harvard School of Public Health (2019) and the same WHO (2011), T2DM is

largely preventable. Its treatment, nonetheless, has an ever-increasing cost and poses a serious threat to future health budgets (Diabetes Canada 2009). Also, prevention must aim at acting on problems before morbidities have the potential to occur.

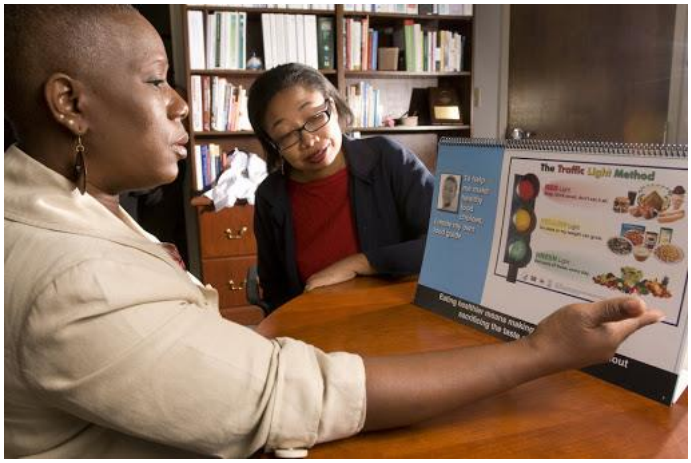
For these reasons, the present review is aimed at addressing the extent to which identifying prediabetes in adults of ≥40 years of age – or younger if presenting with a risk factor according to CDC and Diabetes Canada (2019) – within the HbA1C interval of 5.7-6.4%, as compared to 6.0-6.4%, prevents or delays the onset of Type 2 Diabetes Mellitus, as well as the extent to which it prevents or delays the onset of early symptoms of Type 2 Diabetes Mellitus.

Methods

To ensure the quality of this systematic review, we have completed and published its relative protocol on the PROSPERO database for systematic reviews (use registration ID CRD42019134344 to locate protocol available on the web address: www.crd.york.ac.uk/prospero).

Literature Search and in-exclusion criteria

The databases searched were PubMed, Embase, and the Cochrane Library. A string search sample is provided in the appendix to this article. Initially, the criteria for inclusion allowed only for



human randomized controlled trials (RCTs), published between January 1st, 2008 and January 1st, 2018, in either English, French, Hebrew, Italian, and Spanish, concerning the prevention or delay of pre-diabetes in adults ≥40 years of age, and adopting the HbA1C as a marker. Studies carried out in countries where labs do not adhere to IFCC-WG calibration standards for HbA1C and studies primarily considering patients/participants with other medical conditions or co-morbidities were excluded. Also, a search for grey literature was conducted via searching the PROSPERO database for relevant research proposal submissions, the websites of the department of health of the USA, Canada, and the United Kingdom, and by direct networking with primary care physicians in South Calgary. This search yielded one relevant result – a report from the National Screening

Committee to the National Institute for Clinical Excellence of the Department of Health of the British Government.

First protocol inclusion modification

RCTs were initially the only type of studies contemplated in the inclusion criteria because they would not pose the challenge of possible bias in the population stratification of each study. Given, however, the lack of relevant RCTs found on the databases searched, the entire team agreed to include any clinical trial in the criteria and to ascertain the validity of the methods used by the authors of each article to minimize selection bias in the stratification of the population.

Other methods were also employed to identify relevant research, such as contacting Diabetes Canada and the American Diabetes Association, with the last one being the only agency that replied, and promptly, to our email, while the Canadian one ignored and even declined our multiple requests for information by both email and telephone respectively.

In order to further assure good standards of review practice, two main reviewers were employed to carry out the literature searches, and review of study quality, Dr. Dawid Karczewski (DK) and Dr. Tomasz Karczewski (TK), and a third one, Dr. Mihaela Olsen (MO), was employed for any possible disagreement concerning any study by the initial two reviewers. The data extraction was performed by DK.

Second protocol inclusion modification and final data abstraction

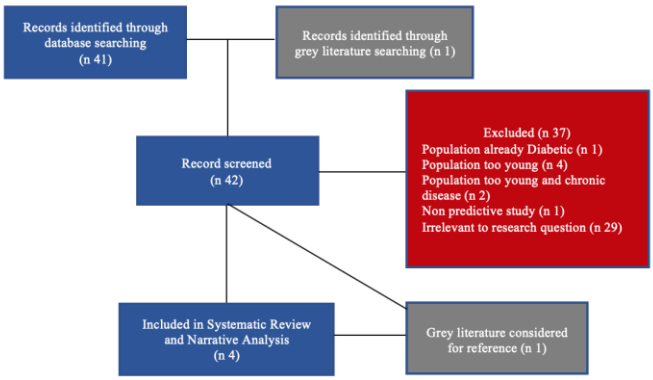
With the new modifications in place to retrieve relevant publications, a total f 41 papers were identified. Still, only 1 cohort study included exclusively patients ≥40 years of age. Other 3, included participants ≥18. It was, therefore, agreed by the team that since CDC (2019) and Diabetes Canada (2019) both recommend testing those below 40 years of age (if they present with at least one predisposing factor) to include also those initially excluded studies, provided that these factors were only hypertension, obesity, and hypercholesterolemia, and that no co-morbidity had already been detected in those patients due to these factors. After these changes, 2 publications were agreed upon by DK and TK. Other 2 remained disputed. MO was presented with the two studies (a systematic review and a prospective study) and with the rationales offered by the two main reviewers for and against their inclusion, and after a thorough examination, MO voted in favor of both. This brought the number of acceptable publications to a final total of four: 1 systematic review of cohort studies, 1 cross-sectional study, and 2 cohort studies (Figure 1).

Even though the need for modification of the inclusion criteria was evidently necessary, the team, by agreeing on the

type of data, as well as terminology prior to the actual review, safeguarded the validity of the modifications of the protocols, and thus the relevance of the data extracted to the original questions of the review itself.

The GRADE approach was applied to rate the quality of the body of evidence being used in this systematic review. The data extraction process was done on a Word document and Zotero 5.0.60 was used to keep account of all references.

Figure 1. Flow chart showing study selection



Data Analysis and Narrative Synthesis

Characteristics and Qualities

Given the heterogeneity of the studies identified concerning their design, geographical settings, research questions, and primary aims, it was impossible to aggregate or pool the data. This was reflected also in the assessment of the quality of their data, which was carried out in a study-by-study fashion using the GRADEpro software free online version from McMaster University, Canada. At the same time, the Critical Appraisal Skills Programme checklist for observational studies/cohort studies was utilized, and its critical content integrated into the GRADE tables in the notes section. This was done to further motivate each assessment carried out with the help of the GRADE approach (Table 5).

Quality Assessment

The certainty assessment, for each study, looked into the design, risk of bias, inconsistency, indirectness, and imprecision. One of the major weak points, for three of the four studies, was the indirectness of the research question. The only study that proposed precise and comparable outcomes in its question, presented a problem of indirectness due to the difference in the population. As a result, the quality of all of the articles in exam was considered to be seriously affected by indirectness, thus resulting in the decision to utilize the narrative template of the GRADE software to report and critically analyze the data from each study.

Starting with the lowest scoring from Selvin et al. (2010), the study was found to be the most open to critically low levels of certainty. Like all others, it was an observational study, which in the hierarchy of evidence would not be

placed in a position of strength. However, it is very unlikely that studies calculating the risk of an event to occur in the long term may be randomized control trials. The very serious risk of bias, nonetheless, came from the very design of the study, which relies on one single HbA1C measurement at baseline, as well as a complete lack of validation of self-reported diabetes diagnosis for the 15-year analysis. This poses the greatest risks for information bias, that cannot be addressed in any sufficient way throughout the study. Also, residual bias cannot be excluded. Further on, the authors suggest that CVD, which is often due to diabetes (Di Pino et al. 2017 and Waugh et al. 2013), may be identified at HbA1C of 5.5% but then, they continue by affirming that an HbA1C of 6.0% is "useful to identify those both at risk of developing T2DM and CVD events". This highlights a major reason for concern for consistency with the results and with the very rationale for the study itself (Table 1).

The second lowest-scoring study, that of Ackerman et al. (2011), is structured and designed according to a complex cohort/quasi-experimental fashion, where no control group is present but a sub-group from the cohort itself is examined to compare the results in the US general population sample and the results if the sample included only those who fall under the CDC risk factors group. This last point strengthens the results of this study concerning the original research question of this very systematic review.

The population was clearly defined as the adult US population. The cohort was recruited in a way that is representative of the US population with stratified, multistage probability sampling with oversampling of older people and minority groups through US National Health and Nutrition Examination Survey (2005-2006). The pre-set in/exclusion criteria were diligently outlined and

observed, and the composite risk was calculated on different A1C levels. To show the beneficial effect of the intervention, the composite factor is identified with levels of HbA1C, and by identifying those at risk who would benefit from early intervention. Measures to minimize possible selection and information bias were put in place. For example, applying to all subjects of the study the same in/exclusion criteria and procedures and by adopting a standardized stratification sampling strategy.

Also, the A1C measurements respected international calibrations protocols. One standard method was adopted to measure A1C in all participants, while the Stern-and-colleagues method was used to estimate the average probability (composite risk) of developing T2DM after a mean of 7.5 years. Sample weights were used in all analysis for the probability of being sampled, for non-coverage, and non-response. Reduction of lead-time bias was accounted for by screening all participants for possible unknown diabetes using the same

standard on selection. All these measures are put in place to reduce the risk of bias. The confidence intervals, however, were not reported. This affects greatly the credibility of the results (Table 2).

The most up-to-date study considered for this review, that of Heianza et al. (2012), is also the only one that expresses precision in its multi-outcome research question. As already expressed at the beginning of the section, however, the results may be applied primarily to Japanese and East Asian individuals. It is therefore unknown if they may apply to a wider setting, such as all other first world countries, including the USA and Canada. Risks for imprecision, in particular, but also inconsistency and bias were kept to a minimum by a rigorous selection process, a standardized initial screening for all participants, and the consideration for a vast number of possible variables (Table 3).

Finally, the systematic review from Zhang et al. (2010), is the highest-scoring study on the certainty assessment of the GRADE tool. The only major weakness of this publication is given by the indirectness of the research question, which does not address two comparable outcomes. Instead, it prospectively looks for the HbA1C threshold concerning an increase in risk in the future development of T2DM (Table 4).

Results

Statement of Principal Findings

Based on the in-exclusion criteria set out in the protocol and their subsequent approved modifications, four studies were identified: three observational studies and a systematic review of observational studies. Due to the heterogeneity of their research questions, design, geographical regions, and methods of data collection, there was no meaningful way to aggregate or pool the data and results. For this reason, a narrative synthesis approach was employed. The data quality and the results were screened using the GRADE approach, integrating for each study the observations obtained via the utilization of the CASP checklist for the relevant type of observational study in exam.

Three out of four total studies considered, suggest that an HbA1C adopting the range of 6.0-6.4% is highly predictive of future T2DM and that the chances of reversion to normoglycemia are severely improbable. One of these three studies strongly suggests that an HbA1C ranging between 5.7-6.4% is the most likely to yield 50% chances of future reversion to normoglycemia.

The remaining two of this group, instead, suggest that a minimum cut off level of 5.5% should be used to identify those at risk for T2DM for them to increase their chances of reversion to normoglycemia in the short and medium-term. One of these

these two studies, however, agrees that if considering the adult population identifiable as at risk of developing T2DM according to the CDC – and therefore also according to Diabetes Canada due to high similarity in the criteria – the ideal minimum cut-off level for HbA1C should be 5.7%.

The final paper instead suggests that 6.0% is the ideal minimal cut-off point, even though it is open to the strongest risk of information bias among all the studies examined in this review. The data collected relatively strongly suggests that between the choice of considering 6.0-6.4% and 5.7-6.4% as the ideal HbA1C range for identifying pre-diabetic adults, the latter should be adopted, as that of 6.0-6.4% only helps to predict an almost sure future development of T2DM, virtually precluding any hope for reversion to a normoglycemic state.

Discussion and Conclusion

Results in the context of previous studies

Previous publications, and protocols – apart from the American Diabetic Association have failed to address any precise advice for a clear minimum cut-off level motivating their decision. A very elevated amount of studies available, however, has tried to ascertain the validity of either of the two minimum thresholds (5.7% vs 6.0%) by comparing the levels of HbA1C with concomitants measurements of FPG (Fasting Plasma Glucose). However, all these studies are exposed to both selection and information bias, as they do not consider the possibility that HbA1C may show variations in different ethnic groups or different geographical regions, and at the same time, that FPG is a test that poses an elevated number of information bias due to its reliance on patients compliance and individual trustworthiness.

Strengths and Weaknesses, Clinical Implications, and Areas of Further Research

This paper, however, should be considered the first step in systematically reviewing the available published evidence regarding the level of HbA1C that is mostly to be associated with a clinically sound, considerable risk of development of T2DM, with a vision on preventing such development.

Given the moderate to very-low quality of the data examined in this systematic review, the evidence produced cannot be considered indisputable. Therefore, even though all evidence points to the fact that an HbA1C of 6.0% as a minimum threshold is no longer to be considered viable if of HbA1C is to be intended a preventive measure, it still does not provide a precise minimum threshold between 5.5% and 5.7%. Because of these partially inconclusive results, it would be best to build on this systematic review and utilize these outcomes as a call to produce new, more up-to-date, higher quality, harmonized, and standardized observational data that may take into consideration increased comparability of outcomes.

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Selvin E, Steffes M W, Zhu H, Matsushita K, Wagenknecht L, Pankow J, Coresh J, Brancati F L. (2010) "Glycated Hemoglobin, Diabetes, and Cardiovascular Risk in Nondiabetic Adults". New England Journal of Medicine, March 4; 362(9): 800–811.

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Zhang X, Gregg EW, Williamson DF, Barker LE, Thomas W, McKeever Bullard K, Imperatore G, Williams DE; Albright AL (2010) "A1C Level and Future Risk of Diabetes: A Systematic Review". Diabetes Care, July 33:7, 1665-1673.

String Search Sample (PUBMED)

("Prediabetic State"[Mesh] OR prediabet*[tw]) AND ("Glycated Hemoglobin A"[Mesh] OR HbA1C[tw]) AND diagnosis AND (cut-off OR cutoff OR level* OR range OR value*) AND (prevent* OR delay)

Table 1. Table of Results according to the GRADEpro Quality of Data and Certainty of Results Incorporating the CASP Criteria – Zhang et al. (2010)

Question: What are the Incidence Risks of future Type 2 Diabetes Mellitus associated with different HbA1C thresholds over a period of 5 years?
Setting: Primary Care
Bibliography: X. ZHANG; E. W. GREGG; D. F. WILLIAMSON; L. E. BARKER; W. THOMAS; K. MCKEEVER BULLARD; G. IMPERATORE; D. E. WILLIAMS; A. L. ALBRIGHT (2010) "A1C Level and Future Risk of Diabetes: A Systematic Review". Diabetes Care, July 33:7, 1665-1673.

Certainty assessment							Impact	Certainty	Importance
№ of studies	Study design	Risk of bias	Inconsistency	Indirectness	Imprecision	Other considerations			
Risk of Developing Type 2 Diabetes Mellitus [HbA1C 6.0% or more] (follow up: 5 years; assessed with: Incidence Risk %)									
1	observational studies	not serious	not serious	serious [a]	not serious	very strong association	An HbA1C of 6.0% or higher, yields a VERY HIGH RISK of developing T2DM within 5 years (25-50% Incidence Risk over a period of 5 years) and a relative risk 20 times higher when compared with an HbA1C of <5%. The population size was 26563 individuals (from an initial 44203 pool).	⨁⨁⨁◯ MODERATE	CRITICAL
Risk of Developing Type 2 Diabetes Mellitus [HbA1C 5.5-6.0%] (follow up: 5 years; assessed with: Incidence Risk %)									
1	observational studies	not serious	not serious	serious [a]	not serious	very strong association	An HbA1C 5.5-6.0% yields a substantial risk of developing T2DM (9-25% Incidence Risk over a period of 5 years). The population size was 26563 individuals (from an initial 44203 pool).	⨁⨁⨁◯ MODERATE	CRITICAL

Explanations

a. The authors care to calculate the level at which the risk of developing T2DM substantially increases, therefore, they do not compare two different managements (for example 5.7 vs 6.0). Instead, they calculate this as if they did not know which level marks the minimum threshold for the risk of developing diabetes, which should be used to diagnose pre-diabetes.

Table 2. Table of Results according to the GRADEpro Quality of Data and Certainty of Results Incorporating the CASP Criteria – Heianza et al. (2012)

Question: Which combination of thresholds among [HbA1C 5.7-6.4% + FPG 5.6-6.9%] OR [HbA1C 6.0-6.4% + FPG 5.6-6.9%] OR [HbA1C 5.7-6.4% + FPG 6.1-6.9] OR [HbA1C 6.0-6.4% + FPG 6.1-6.9%] determins a higher risk of developing T2DM in the future?
Setting: Primary Care
Bibliography: Heianza, Y; Arase, Y; Fujihara, K; Tsuji, H; Saito, K; Hsieh, SD; Kodama, S; Shimano, H; Yamada, N; Hara, S; Sone, H. (2012) "Screening for pre-diabetes to predict future diabetes using various cut-off points for HbA(1c) and impaired fasting glucose: the Toranomon Hospital Health Management Center Study 4". Diabetes Medicine, Sep;29(9):e279-85.

Certainty assessment							Impact	Certainty	Importance
№ of studies	Study design	Risk of bias	Inconsistency	Indirectness	Imprecision	Other considerations			
Risk of Developing T2DM in Japanese Adult Population with threshold [HbA1C 5.7-6.4% + FPG 5.6-6.9%] (follow up: 4.5 years; assessed with: Percentage)									
1	observational studies	not serious	not serious	serious [a]	not serious	strong association	A combination of A1C 5.7-6.4% + FPG 5.6-6.9% yields 50% cumulative risk of developing T2DM. After Cox analysis for multivariate responsible for reversion yields 32.5% Hazard Risk (C.I. 95% range 23.0-45.8). The population size was 6241 individuals (from an initial pool of 6636).	⨁⨁◯◯ LOW	CRITICAL
Risk of Developing T2DM in Japanese Adult Population with threshold [HbA1C 6.0-6.4% + FPG 5.6-6.9%] (follow up: 4.5 years; assessed with: Percentage)									
1	observational studies	not serious	not serious	serious [a]	not serious	strong association	A combination of A1C 6.0-6.4% + FPG 5.6-6.9% yields 80% cumulative risk of developing T2DM. After Cox analysis for multivariate responsible for reversion yields 53.7% Hazard Risk (C.I. 95% range 38.4-75.1). The population size was 6241 individuals (from an initial pool of 6636).	⨁⨁◯◯ LOW	CRITICAL
Risk of Developing T2DM in Japanese Adult Population with threshold [HbA1C 5.7-6.4% + FPG 6.1-6.9%] (follow up: 4.5 years; assessed with: Percentage)									
1	observational studies	not serious	not serious	serious [a]	not serious	strong association	A combination of A1C 5.7-6.4% + FPG 6.1-6.9% yields 80% cumulative risk of developing T2DM. After Cox analysis for multivariate responsible for reversion yields 37.9% Hazard Risk (C.I. 95% range 28.18-51.1). The population size was 6241 individuals (from an initial pool of 6636).	⨁⨁◯◯ LOW	CRITICAL
Risk of Developing T2DM in Japanese Adult Population with threshold [HbA1C 6.0-6.4% + FPG 6.1-6.9%] (follow up: 4.5 years; assessed with: Percentage)									
1	observational studies	not serious	not serious	serious [a]	not serious	strong association	A combination of A1C 6.0-6.4% + FPG 6.1-6.9% yields 100% cumulative risk of developing T2DM. After Cox analysis for multivariate responsible for reversion yields 52.3% Hazard Risk (C.I. 95% range 37.8-72.3). The population size was 6241 individuals (from an initial pool of 6636).	⨁⨁◯◯ LOW	CRITICAL

CI: Confidence interval; **HR:** Hazard Ratio

Explanations

a. The Japanese population is ethnically very uniform. Therefore, as also suggested by the authors, the results may be applicable primarily to those of Japanese and East Asian origin.

Table 3. Table of Results according to the GRADEpro Quality of Data and Certainty of Results Incorporating the CASP Criteria – Ackermann et al. (2011)

Question: What is the probability of developing Type 2 Diabetes Mellitus over a 7.5-year period for every different HbA1C threshold?
Setting: Primary Care
Bibliography: Ackermann RT, Cheng YJ, Williamson DF, Gregg EW (2011). "Identifying adults at high risk for diabetes and cardiovascular disease using hemoglobin A1c National Health and Nutrition Examination Survey 2005-2006". American Journal of Preventive Medicine 40:11–17

Certainty assessment							Impact	Certainty	Importance
№ of studies	Study design	Risk of bias	Inconsistency	Indirectness	Imprecision	Other considerations			
Risk of Developing Type 2 Diabetes Mellitus - IN STANDARD U.S. POPULATION SAMPLE (follow up: mean 7.5 years; assessed with: Percentage)									
1	observational studies	serious [a]	not serious	serious [b]	not serious	strong association	A HbA1C of 5.5% and above is suitable for defining pre-diabetes in the general adult population. The population size was 1798 individuals (from an initial 4751 pool).	⊕◯◯◯ VERY LOW	IMPORTANT
Risk of Developing Type 2 Diabetes Mellitus - IN U.S. POPULATION SAMPLE WITH DPP CRITERIA (follow up: mean 7.5 years; assessed with: Percentage)									
1	observational studies	serious [a]	not serious	serious [b]	not serious	strong association	A HbA1C of 5.7% and above is appropriate for those who meet the DPP enrolment criteria. The population size was 1798 individuals (from an initial 4751 pool).	⊕◯◯◯ VERY LOW	IMPORTANT

Explanations

a. The confidence interval was not reported. This affects greatly the credibility of the results
b. The authors care to calculate the level at which the risk of developing T2DM substantially increases, therefore, they do not compare two different managements (for example 5.7 vs 6.0). Instead, they calculate this as if they did not know which level marks the minimum threshold for the risk of developing diabetes, which should be used to diagnose pre-diabetes.

Table 4. Table of Results according to the GRADEpro Quality of Data and Certainty of Results Incorporating the CASP Criteria – Selvin et al. (2010)

Question: What are the Hazard Ratios for different HbA1C thresholds for developing diabetes within a 15 years period?
Setting: Primary Care
Bibliography: Selvin E, Steffes M W, Zhu H, Matsushita K, Wagenknecht L, Pankow J, Coresh J, Brancati F L. (2010) "Glycated Hemoglobin, Diabetes, and Cardiovascular Risk in Nondiabetic Adults". New England Journal of Medicine, March 4; 362(9): 800–811.

Certainty assessment							Impact	Certainty	Importance
№ of studies	Study design	Risk of bias	Inconsistency	Indirectness	Imprecision	Other considerations			
Risk of Developing Type 2 Diabetes Mellitus [HbA1C less than 5.0%] (follow up: 15 years; assessed with: Hazard Ratio)									
1	observational studies	very serious [a]	serious [b]	serious [c]	serious [b]	strong association	Hazard Ratio 0.52 (95% C.I. interval 0.40-0.69). The population size was 11092 individuals (from an initial 15792 pool).	⨁◯◯◯ VERY LOW	IMPORTANT
Risk of Developing Type 2 Diabetes Mellitus [HbA1C 5.0 - 5.5%] (follow up: 15 years; assessed with: Hazard Ratio)									
1	observational studies	very serious [a]	serious [b]	serious [c]	serious [b]	strong association	Hazard Ratio 1.00 (reference). The population size was 11092 individuals (from an initial 15792 pool).	⨁◯◯◯ VERY LOW	IMPORTANT
Risk of Developing Type 2 Diabetes Mellitus [HbA1C 5.5 - 5.9%] (follow up: 15 years; assessed with: Hazard Ratio)									
1	observational studies	very serious [a]	serious [b]	serious [c]	serious [b]	strong association	Hazard Ratio 1.86 (95% C.I. interval 1.67-2.08). The population size was 11092 individuals (from an initial 15792 pool).	⨁◯◯◯ VERY LOW	IMPORTANT
Risk of Developing Type 2 Diabetes Mellitus [HbA1C 6.0 - 6.4%] (follow up: 15 years; assessed with: Hazard Ratio)									
1	observational studies	very serious [a]	serious [b]	serious [c]	serious [b]	strong association	Hazard Ratio 4.48 (95% C.I. interval 3.92-5.13). Identified by Authors as "useful to identify those both at risk of developing T2DM and CVD events".	⨁◯◯◯ VERY LOW	IMPORTANT
Risk of Developing Type 2 Diabetes Mellitus [HbA1C 6.5 or more] (follow up: 15 years; assessed with: Hazard Ratio)									
1	observational studies	very serious [a]	serious [b]	serious [c]	serious [b]	strong association	Hazard Ratio 16.47 (95% C.I. interval 14.22-19.08). The population size was 11092 individuals (from an initial 15792 pool).	⨁◯◯◯ VERY LOW	IMPORTANT

CI: Confidence interval

Explanations

a. The very serious risk of bias came from the very design of the study, which relies on one single HbA1C measurement at baseline, as well as a complete lack of validation of self-reported diabetes diagnosis for the 15-year analysis. This poses the greatest risks for information bias, that cannot be addressed in any sufficient way throughout the study. In addition, residual bias cannot be excluded.
b. The authors suggest that CVD (which is often due to diabetes) may be identified at HbA1C of 5.5% state that an HbA1C of 6.0% is "useful to identify those both at risk of developing T2DM and CVD events". This poses a concern for consistency with results.
c. The authors care to calculate the level at which the risk of developing T2DM substantially increases, therefore, they do not compare two different managements (for example 5.7 vs 6.0). Instead, they calculate this as if they did not know which level marks the minimum threshold for the risk of developing diabetes, which should be used to diagnose pre-diabetes.

Table 5. Condensed Table of Results according to the GRADEpro Quality of Data and Certainty of Results Incorporating the CASP Criteria

Study	Inconsistency	Imprecision	Risk of Bias	Indirectness	Study Population Size	Certainty	Minimum HbA1c level proposed for identifying adult pre-diabetics
Zhang et al (2010)	NO	NO	NO	YES (a)	**26563** (from initial 44203)	MODERATE	5.5-6.0%
Heianza et al (2012)	NO	NO	NO	YES (b)	**6241** (from initial 6636)	LOW	5.7-6.4% but only together with FPG of 5.6-6.9%
Ackermann et al (2011)	NO	NO	YES Serious (e)	YES (a)	**1798** (from initial 4751)	VERY LOW	5.5-6.4% OR 5.7-6.4% in those at risk (CDC Criteria)
Selvin et al (2010)	YES Serious (c)	YES Serious(c)	YES Very Serious (d)	YES (a)	**11092** (from initial 15792)	VERY LOW	6.0-6.4%

LEGEND

a. The authors care to calculate the level at which the risk of developing T2DM substantially increases, therefore, they do not compare two different managements (for example 5.7 vs 6.0). Instead, they calculate this as if they did not know which level marks the minimum threshold for the risk of developing diabetes, which should be used to diagnose pre-diabetes.

b. The Japanese population is ethnic ally very uniform. Therefore, as also suggested by the authors, the results may be applicable primarily to those of Japanese and East Asian origin.

c. The authors suggest that CVD (which is often due to diabetes) may be identified at HbA1C of 5.5% but state that an HbA1C of 6.0% is "useful to identify those both at risk of developing T2DM and CVD events". This poses a concern for consistency with the results.

d. The study's reliance on one single HbA1C measurement at baseline, as well as the lack of validation of self-reported diabetes diagnosis for the 15-year analysis, pose the greatest risks for information bias, that cannot be addressed in any sufficient way throughout the study. Residual bias cannot be excluded.

e. The confidence interval was not reported. This affects greatly the credibility of the results.