The new diagnostic threshold of hemoglobin A1c was made based on evidence from cross-sectional studies, and no longitudinal study supports its validity. To examine whether hemoglobin A1c of 6.5% or higher defines a threshold for elevated risk of incident retinopathy, we analyzed longitudinal data of 19,897 Japanese adults who underwent a health checkup in 2006 and were followed up 3 years later. We used logistic regression models and restricted cubic spline models to examine the relationship between baseline hemoglobin A1c levels and the prevalence and the 3-year incidence of retinopathy. The restricted cubic spline model indicated a possible threshold for the risk of incident retinopathy at hemoglobin A1c levels of 6.0–7.0%. Logistic regression analysis found that individuals with hemoglobin A1c levels of 6.5–6.9% were at significantly higher risk of developing retinopathy at 3 years compared with those with hemoglobin A1c levels of 5.0–5.4% (adjusted odds ratio, 2.35 [95% CI 1.08–5.11]). Those with hemoglobin A1c levels between 5.5 and 6.4% exhibited no evidence of elevated risks. We did not observe a threshold in the analysis of prevalent retinopathy. Our longitudinal results support the validity of the new hemoglobin A1c threshold of 6.5% or higher for diagnosing diabetes. Diabetes 61:3280–3284, 2012

Diabetes is an increasingly important global public health concern (1). An estimated 285 million people, or 6.4% of the world’s population, lived with diabetes in 2010, and the number is expected to grow to 438 million by 2030 (1). In the U.S., 8.3% of children and adults are living with diabetes (2); likewise, in Japan, 7.8% of the population has diabetes (3).

Recently, the International Expert Committee suggested use of a hemoglobin A1c (HbA1c) level of 6.5% or higher as the threshold for diagnosing diabetes (4,5). This criterion was subsequently adopted by the American Diabetes Association, European Association for the Study of Diabetes, and World Health Organization (4,5). In making its decision, the expert panel was informed by evidence from several cross-sectional studies that showed the association between HbA1c level and the prevalence of retinopathy (4–12). The outcome of retinopathy has been historically accepted as the best criterion for comparing glycemic measures among several complications of diabetes (13,5), because it is a specific complication of diabetes that can be measured objectively (13,14). Few longitudinal studies have examined the association between HbA1c levels and the risk of retinopathy in the general population, and these studies do not support the validity of this new diagnostic threshold (6,15–17). Many of the previous studies did not adjust for independent risk factors and confounders for retinopathy, such as age and hypertension.

To examine the validity of the new HbA1c thresholds, we tested the hypothesis that HbA1c level of 6.5% or higher would define a threshold for increased 3-year incidence of retinopathy in a large cohort of Japanese adults.
RESULTS

Study participants. Table 1 shows the baseline characteristics of the study population overall and according to HbA1c levels. Approximately 49% were men. The mean age was 51.0 years, the mean HbA1c was 5.6%, and the mean (±SD) follow-up period was 3.0 ± 0.29 years. Participants with higher HbA1c levels were more likely to be older, to be men, to smoke, to take medication for hypertension, and to have several clinical risk factors, including elevated blood pressure, higher BMI, lower HDL cholesterol, higher triglyceride, and a family history of diabetes.

Prevalence of retinopathy. Among the 20,433 participants, the crude prevalence of retinopathy was 1.2% (245/20,433). The adjusted ORs and 95% CIs for prevalent retinopathy are shown in Table 2. After initial adjustment for age and sex (model 1a), the prevalence of retinopathy was significantly higher at HbA1c levels ≥6.5% than in the reference category. After further adjustments (model 2a), this estimate was attenuated, and only HbA1c levels ≥7.0% achieved statistical significance.

Cumulative incidence of retinopathy. The crude cumulative incidence of retinopathy at 3 years was 0.85%...
(170/19,897). After initial adjustment for age and sex, there was no significant association between HbA1c value and the incidence of retinopathy at HbA1c <6.5% compared with the reference category, however, HbA1c levels of 6.5–6.9% were associated with significantly higher risk of developing retinopathy at 3 years (OR 2.35 [95% CI 1.08–5.11]; P = 0.031) (Table 3). The risk remained significantly higher after further adjusting for the confounders and other independent risk factors for retinopathy (model 2b). Our results did not alter substantially after the exclusion of those with diagnosis of diabetes at baseline in model 3b.

In our analysis to evaluate whether the relationship between baseline HbA1c levels and prevalence of retinopathy is nonlinear, we found that the nonlinear relationship was significant (P for curve = 0.08) (Fig. 1). For the outcome of incident retinopathy, however, the nonlinear relationship was statistically significant (P = 0.001), suggesting a possible threshold around HbA1c levels between 6.0 and 7.0%. We observed a dose-response relationship between higher HbA1c levels and increased risk of incident retinopathy at HbA1c ≥6.5%. The restricted cubic spline analysis for the non-diabetic subpopulation yielded similar findings (data not shown).

### DISCUSSION

We found that Japanese adults with HbA1c levels of 6.5–6.9% were at significantly higher risk of developing retinopathy at 3 years than were those with HbA1c levels of 5.0–5.4%, whereas the risks did not increase among those with HbA1c levels <6.5%. To the best of our knowledge, this is the first longitudinal study supporting the validity of the new diagnostic threshold of HbA1c recommended by the International Expert Committee (5,11). In contrast, we did not observe an explicit threshold effect of HbA1c in the analyses of the prevalent retinopathy.

Although there have been several longitudinal studies examining the association between HbA1c levels and the risk of retinopathy, most of those studies were limited to samples treated for diabetes (29,34–38). There have been only a few small longitudinal studies of general non-diabetic populations (6,15–17). Possibly because of small sample sizes, results from these previous studies did not support the current HbA1c threshold for diagnosing diabetes (6,15–17). In the Pima Indian study (N = 927), investigators found that the risk of incident retinopathy increased only at HbA1c levels of 9.1% (6). Only HbA1c data were available at that time (HbA1c measurements were unavailable), and no adjustments were made for hypertension and other independent risk factors for retinopathy in that study (6). The longitudinal study (N = 233) by van Leiden et al. (15) found that the risk of developing retinopathy was significantly higher in the highest tertile of HbA1c (HbA1c 5.8–13.1%) relative to the lowest tertile (HbA1c 4.3–5.2%). Because of small sample size, these investigators collapsed HbA1c levels from 5.8 to 13.1% into a single category. Selvin et al. (16) examined the risk of retinopathy among participants from the Atherosclerosis Risk in Communities study. The overall sample size of this study was large (N = 11,357), but repeated retinal examinations were performed in only 767 people. These investigators were unable to detect a statistically significant association with HbA1c value.

![Table 2](https://example.com/table2.png)

**Table 2**

Adjusted ORs and 95% CIs for the prevalence of retinopathy across different baseline HbA1c levels

| HbA1c (%) | N   | No. of cases (%) | OR  | 95% CI   | P      | Model 1a* | N   | No. of cases (%) | OR  | 95% CI   | P      | Model 2a† |
|----------|-----|------------------|-----|---------|--------|-----------|-----|------------------|-----|---------|--------|-----------|
| <5.0     | 1,719 | 10 (0.6)  | 1.17 | 0.60–2.29 | 0.65   | 1.08     | 0.55–2.12 | 0.83 | 1,692  | 10 (0.6)  | 1.20     | 0.61–2.36 | 0.60  |
| 5.0–5.4  | 9,300 | 67 (0.7)   | 1.00 | Ref.    | Ref.   | 1.00     | Ref.     | Ref. | 9,170  | 65 (0.7)  | 1.00     | Ref.     | Ref.   |
| 5.5–5.9  | 6,376 | 76 (1.2)   | 1.14 | 0.81–1.59 | 0.46   | 1.16     | 0.82–1.63 | 0.40 | 6,188  | 72 (1.2)  | 1.12     | 0.79–1.58 | 0.54  |
| 6.0–6.4  | 2,046 | 26 (1.3)   | 0.91 | 0.57–1.46 | 0.71   | 0.88     | 0.54–1.42 | 0.59 | 1,893  | 21 (1.1)  | 0.80     | 0.48–1.34 | 0.39  |
| 6.5–6.9  | 416   | 17 (4.1)   | 2.63 | 1.50–4.59 | 0.0007 | 1.81     | 0.98–3.37 | 0.060| 268    | 5 (1.9)   | 1.18     | 0.46–3.03 | 0.73  |
| ≥7.0     | 576   | 49 (8.5)   | 5.69 | 3.82–8.47 | <0.0001| 3.02     | 1.71–5.34 | <0.0001| 130    | 5 (3.9)   | 3.00     | 1.14–7.84 | 0.026 |

Statistically significant results at P < 0.05 are indicated in boldface. *Adjusted for age and sex. †Adjusted for age, sex, hypertension, diagnosis of diabetes, HDL and LDL cholesterol, log-transformed triglyceride, BMI, alcohol consumption, smoking status, and family history of diabetes. ‡Adjusted for all the variables in model 2a, with the exception of the diagnosis of diabetes.
significant threshold in the association of HbA1c with the incidence of retinopathy, possibly because of the lack of power. Recently, Massin et al. (17) studied 700 participants from the Data from an Epidemiological Study on the Insulin Resistance Syndrome study, in which participants were followed up for 10 years, and proposed an HbA1c threshold of 6.0%. Because retinopathy was not evaluated at baseline in this study, however, they were not able to examine the incidence of retinopathy (17).

Our study has several limitations. First, the retinal images were graded in single-field photographs per eye in this study. Multiple photographic fields per eye would have improved the sensitivity of the funduscopic examinations. Second, our study sample was composed exclusively of native Japanese, so whether our results generalize to other populations is unclear. It is noteworthy that in the DETECT-2 project, which pooled studies from the U.S., Australia, India, Japan, and Singapore, they found no racial difference in optimal HbA1c threshold (12). Third, we did not take into account the possible effect of hemoglobinopathies on HbA1c values. However, the prevalence of hemoglobinopathies in Japan is reported to be as low as 0.04% (39) and is therefore likely to have little impact on our overall findings. Finally, the detection of an inflection point in the relation between HbA1c and retinopathy may not in itself establish the optimal threshold for clinicians to use in the diagnosis and treatment of diabetes. The optimal threshold for any patient is the level at which the benefits of diagnosis and treatment exceed harms for that patient. If there were only benefits and no harm in diagnosing and treating diabetes, the inflection point would represent the level of HbA1c. However, when the benefits of diagnosis and treatment of diabetes are small, the optimal diagnostic threshold may be higher than the inflection point we observed.

Our longitudinal study is the first to date to suggest a threshold of risk for incident retinopathy at a 6.5% HbA1c level. These findings support the validity of the new diagnostic HbA1c threshold for diabetes recently adopted by the American Diabetes Association, the European Association for the Study of Diabetes, and the World Health Organization (5,12). Additional longitudinal studies are needed to validate these findings in other populations.

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Y.T., O.T., T.F., W.C.T., and C.C.W. developed the study concept, developed the design, and interpreted the data. Y.T., R.B.D., and F.I. conducted statistical analyses. Y.T., J.B.M., W.C.T., and C.C.W. drafted the manuscript. All the authors revised the manuscript critically for important intellectual content and approved the final manuscript. Y.T. is the guarantor of this work and, as such, had full access to all the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis.

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